

European Society for Evolutionary Biology

ORAL PRESENTATION ABSTRACTS AND LIST OF POSTERS

14th Congress, Lisbon, Portugal, 19 - 24 August 2013

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Plenary Lectures

1. Tuesday August 20

Juliette De-Meaux, University of Münster, Germany "The molecular underpinnings of adaptation in *Arabidopsis thaliana*"

2. Wednesday August 21

Dieter Ebert, University of Basel, Switzerland "Population genetics of host-parasite coevolution"

3. Thursday August 22

Mel Greaves, The Institute of Cancer Research, United Kingdom "Cancer through the eye of evolutionary medicine"

4. Friday August 23

Trudy Mackay, North Carolina State University, United States "Charting the Genotype-Phenotype map: Lessons from *Drosophila*"

5. Saturday August 24

Virpi Lummaa, University of Sheffield, United Kingdom "Sex differences in natural selection on reproductive scheduling and longevity in humans"

PRESIDENTIAL ADDRESS

Roger Butlin, University of Sheffield, United Kingdom "Reinforcement (and other modes of speciation)"

JOHN MAYNARD SMITH PRIZE LECTURE

Richard FitzJohn, Macquarie University, Australia "What drives biological diversification? Detecting traits under species selection"

DISTINGUISHED FELLOW LECTURE

Rolf Hoekstra, Wageningen University, Netherlands

"♀ ♂ **+ -**"

Plenary (Tue 20)

THE MOLECULAR UNDERPINNINGS OF ADAPTATION IN ARABIDOPSIS THALIANA

Juliette De-Meaux¹

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The theories used in Evolutionary Biology are rooted in powerful concepts, most of which were constructed before the discovery of DNA. In the meantime, we understand much better how function is encoded in the genome. The current task is now to examine how genetic modifications in molecular systems can promote adaptation in natural populations. *Arabidopsis thaliana* is a focal system in plant molecular biology. Genes and pathways controlling its development, physiology and immunity have been researched intensively. Its broad latitudinal distribution in Europe makes it a unique system to dissect the molecular underpinnings of adaptation. I will review some of the molecular changes that have been successfully associated with adaptive evolution in *A. thaliana*. I will thereby illustrate why reconciling molecular and evolutionary biology is a difficult yet necessary task, requiring a shift of concepts in both molecular biology and evolutionary theory.

Plenary (Wed 21)

POPULATION GENETICS OF HOST-PARASITE COEVOLUTION

Dieter Ebert¹

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Hosts evolve to minimize the fitness reduction caused by parasites, while parasites optimise the exploitation of their hosts. In models of this process high genetic specificity in host – parasite interactions is assumed. These interactions are in the centre of theory of host – parasite coevolution and determine important aspects of the coevolutionary process, such as its tempo and mode, the occurrence of cyclic allele frequencies, and the potential for evolutionary novelty. They are also crucial for the consequences of coevolution, such as the maintenance of sex and of genetic variation. In my presentation I will analyse the genetic architecture of host-parasite interactions and will test predictions and assumptions of models of host – parasite coevolution. Finally I will match predictions with patterns from long-term coevolution records.

Plenary (Thu 22)

CANCER THROUGH THE EYE OF EVOLUTIONARY MEDICINE

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There are two striking examples of somatic cell evolution in humans (and vertebrates in general): the immune system and cancer. In the former, recombinase enzymes target diversification of the immunoglobulin and T cell receptor loci in lymphocytes provides the substrate (>1011 variants) for natural selection by infectious antigenic epitopes. In cancer, stochastic mutational processes drive genetic diversification of clones of cell that under natural selection within complex tissue ecosystems and in response to therapeutic pressure. Starting from a single founder cell, cancer clones evolve over time frames of 1 to 50 years, often in a covert fashion, but the end result is the generation of a robust and weed-like quasi-species of immortal and invasive cells that hijack normal tissue function. In exceptional circumstances, cancer cells transit, parasitic like, individual to individual. Therapy itself inadvertently provides positive selective pressure for the emergence of drug-resistant mutant subclones. Whole genome sequencing reveals the extent of genetic, mutational complexity in cancer and when interrogated at the single cell level, it is clear that cancers have tree-like clonal phylogenies with variegated genetics of sub-clones. Within sub-clones, cells with stem cell properties sustain and propel clonal expansion and provide the cellular units of selection. Cancer's Darwinian character has substantial implications for prevention, prognosis and treatment.

Plenary (Fri 23)

CHARTING THE GENOTYPE-PHENOTYPE MAP: LESSONS FROM DROSOPHILA

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Quantitative traits are affected by multiple interacting loci with individually small and environmentally sensitive effects. Knowledge of the detailed genetic architecture of quantitative traits is important from the perspectives of evolutionary biology, human health and plant and animal breeding. Understanding the genetic architecture of quantitative traits begins with identifying the genes regulating these traits, mapping the subset of genetically varying quantitative trait loci (QTLs) in natural populations, and pinpointing the molecular polymorphisms defining QTL alleles. *Drosophila* brings an impressive toolkit to the challenge of genetically dissecting quantitative traits. I will discuss insights into the complex genetic architecture of quantitative traits obtained from genome wide association mapping in the *Drosophila melanogaster* Genetic Reference Panel (DGRP), which consists of 192 sequenced inbred lines derived from the Raleigh, USA population. These studies indicate that epistatic gene action is common, and additivity can be an emergent property of underlying genetic interaction networks. Epistasis causes hidden quantitative genetic variation in natural populations, the potential for rapid speciation, and negatively impacts the predictive ability of additive models. These observations offer valuable lessons for understanding the genetic basis of variation for quantitative traits in other organisms.

Plenary (Sat 24)

SEX DIFFERENCES IN NATURAL SELECTION ON REPRODUCTIVE SCHEDULING AND LONGEVITY IN HUMANS

Virpi Lummaa¹

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Despite senescence with age, most animals retain ability to reproduce until relatively close to death. Humans provide an interesting case because with mid-life menopause, women show a radical decoupling of senescence in reproductive and somatic systems, leading to up to half of total lifespan spent post-reproductive. By contrast, men maintain reproductive ability until much later ages. Although men thus sire offspring at older ages than women, nearly all contemporary human populations exhibit sexual dimorphism in lifespan with women outliving men by on average of five years. While proximate causes for such fertility and lifespan differences between the sexes are well-known, our understanding of the underlying evolutionary forces is much more limited. I use pedigree data on pre-industrial Finnish men and women collected by local clergymen: (1) to address different evolutionary hypothesis for the benefits of menopause and post-reproductive longevity in women; (2) to assess whether selection on overall lifespan differs between the sexes; (3) and to estimate sex-specific heritabilities for, and genetic correlations between, lifespan and fitness to predict evolutionary trajectories for lifespan and sexual dimorphism. Understanding sex differences in rates of senescence in reproduction and survival, both key life-history traits, provides insights into how differing selection pressures can mould rates of senescence and ultimate longevity within a species. I hope to illustrate that although evolutionary studies on contemporary human populations suffer from many limitations, some of the data available on humans offer interesting research opportunities also for evolutionary biologists with potential implications for studies on demography, public health or anthropology.

Presidential Address

REINFORCEMENT (AND OTHER MODES OF SPECIATION)

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Reinforcement is often considered special among modes of speciation because natural selection favours the build-up of barriers to gene flow: reproductive isolation is not just an incidental consequence of divergence, it is a device to prevent maladaptive hybridization. The idea is controversial partly because the term 'reinforcement' is used in various different ways. More importantly, reinforcement is hard to demonstrate and so it remains uncertain how much it contributes to speciation. Like other processes, it is only ever part of the speciation story. I will discuss the role of reinforcement is association with various other elements of speciation, using evidence from some of my favourite organisms.

JMS Prize

WHAT DRIVES BIOLOGICAL DIVERSIFICATION? DETECTING TRAITS UNDER SPECIES SELECTION.

Richard G FitzJohn¹

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Species selection -- heritable trait-dependent differences in rates of speciation or extinction -- may be responsible for variation in both taxonomic and trait diversity among clades. While initially controversial, interest in species selection has been revived by the accumulation of evidence of widespread trait-dependent diversification. I will present several methods for investigating species selection by detecting the association between species traits and speciation or extinction rates. These methods are explicitly phylogenetic and incorporate simple, but commonly used, models of speciation, extinction, and trait evolution. Using these methods, I will present several examples where traits are correlated with speciation or extinction rates in plants and mammals. All methods have assumptions and limitations, and I will discuss the pitfalls that arise when applying these methods (and the widely used methods that they derive from) to messy biological data. Comparative phylogenetic methods must be used with caution, but allow testing of long-standing hypotheses about causes of variation in biological diversity.

Distinguished Fellow

♀ ♂ **+ -**

Rolf Hoekstra¹

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Over a timespan of some 30 years I have intermittently worked on understanding the origin of sexual asymmetry. I greatly enjoyed it, but the problem remains...

Symposium

1. Experimental Evolution on Empirical Fitness Landscapes

22 and 23 August



Program

Thursday 22 August

Session(s): 9 Friday 23 August Session(s): 10, 11

Organisers: J. Arjan G.M. de Visser and Santiago F. Elena

Invited speakers: Tim Cooper and Joachim Krug

Description:

To arrive at a quantitative understanding of adaptation, we need to identify the factors that determine its dynamics and understand how they do so quantitatively. A prominent recent development is to consider the structure of the fitness landscape and how this determines the outcome of adaptation. Microbial experimental evolution contributes to this development by exploring the structure of real fitness landscapes, either by constructing mutants carrying combinations of observed beneficial mutations, or by studying the contingency of evolution on particular genotypes and conditions. The aim of this symposium is to highlight diverse examples of the empirical study of fitness landscapes using microbial experimental evolution [and their contribution to quantitative models of adaptation].

D22SY01IT10:30R2

EPISTATIC CONSTRAINTS AND EVOLUTIONARY PREDICTABILITY ON EMPIRICAL FITNESS LANDSCAPES

Joachim Krug¹

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The adaptive dynamics of an asexual population in the space of genotypes is constrained by epistatic interactions between mutations at different genetic loci. Recent empirical studies have shown that this strongly reduces the number of mutational pathways that are accessible under conditions of strong selection and weak mutation (SSWM). In the talk I will describe statistical models for fitness landscapes that quantify evolutionary accessibility under different assumptions on the amount of epistasis as well as on the underlying genetic architecture, and show how these models can be used to classify and interpret empirical data sets. I then discuss the impact of epistatic constraints on the predictability of evolutionary trajectories in asexuals, with particular emphasis on the role of population size. With increasing population size clonal interference implies a preference for mutational steps of large effect, which leads to an increase in predictability beyond the expectation under SSWM dynamics. However, a further increase of population size reduces predictability by opening up new pathways that involve the crossing of fitness valleys by multiple mutations. This nonmonotonic pattern of evolutionary predictability is found in large-scale simulations on an empirical fitness landscape, and argued to be observable in experiments that monitor the variability of fitness trajectories among replicate populations.

D23SY01IT10:30R2

EPISTASIS AND EVOLVABILITY IN EXPERIMENTALLY EVOLVED POPULATIONS OF ESCHERICHIA COLI

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Epistatic interactions between mutations play a prominent role in many evolutionary theories. Many studies have found that epistasis is widespread, but direct analyses of epistasis can be technically difficult and has not generally considered beneficial mutations. We analyse the effects of epistasis on fitness in a set of genotypes including all combinations of the first five beneficial mutations to fix in an experimental population of Escherichia coli. We show that epistasis depends strongly on the fitness effects of the combined mutations – the larger the expected benefit, the more negative the effect of epistasis on fitness. A similar pattern of interactions is also seen among a set of seven mutations that substituted in a population that was selected in an environment containing a combination of two sugars. Epistasis thus tended to follow a simple relationship of diminishing returns with genotype fitness. This observation supports a model that predicts negative epistasis explains a decelerating rate of adaptation as populations approach a fitness peak. Preliminary experiments are consistent with this prediction, finding that the effective size, but not rate, of beneficial mutations declines across a series of replay populations started from genotypes of progressively higher fitness.

D22SY01RT11:18R2

IMPACT OF GENE ARCHITECTURE ON THE ADAPTATION RATE

<u>Lilia Perfeito</u>¹, Miguel Godinho Ferreira¹ ¹*Instituto Gulbenkian de Ciência, Portugal* lilia.perfeito@gmail.com

The accumulation of spontaneous beneficial mutations is one of the drivers of survival and adaptation in novel environments. Predicting the fitness effects of mutations and explaining them from a functional point of view are among the major goals of modern evolutionary biology. The rate and effects of spontaneous beneficial mutations are expected to depend on the environment, on demography and on genotype. Different models have been proposed to predict and explain the effects of new mutations from the underlying phenotypes. Here we measure the adaptation rate of fission yeast strains with different genome architectures. These strains have identical gene composition. However, they vary in gene expression and show substantial variation for fitness. By following the course of evolution of neutral markers in populations of these strains, we are able to estimate the rate and distribution of effects of new mutations. We use a variant of Fisher's geometrical model (FGM) which can explain the data, while capturing the functional details of the system. FGM predicts that the adaptation rate is a function of initial fitness, even when these differences come from changes affecting multiple genes, as is the case with genomic rearrangements. Our results, taken together with recent observations in experimental evolution, point to the existence of a single peaked landscape with minute complexities at the microscopy level, but a smooth upward climb on the macroscopic level.

D22SY01RT11:42R2

LANDSCAPE RUGGEDNESS REDUCES GENOMIC PARALLELISM IN EXPERIMENTAL POPULATIONS OF *PSEUDOMONAS*

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The repeatability of adaptive evolution depends on the ruggedness of the underlying adaptive landscape, how fitness varies as a function of phenotype or genotype. Rugged landscapes are thought to promote divergent adaptation, with genotypes evolving towards distinct genotypic and phenotypic solutions determined by the number of available fitness peaks. By contrast, genotypes evolving on a smooth landscape containing a single adaptive peak are expected to converge on a single genotypic and phenotypic solution. Here we evaluate the genomic consequences of adaptation on rugged and smooth landscapes by quantifying the degree of genic parallelism observed following adaptive evolution by genetically distinct starting genotypes of *Pseudomonas fluorescens* evolving on two single carbon substrates, xylose and glucose. Previous work showed that these substrates differ in the number of adaptive solutions available to these genotypes, with xylose being a relatively more rugged landscape than glucose. We find that, consistent with expectation, DNA sequence evolution is less parallel in a rugged, compared to a smooth landscape. Our results suggest that the ruggedness of the adaptive landscape has a strong influence on the pattern of genomic evolution.

D23SY01RT11:18R2

MAPPING THE ADAPTIVE LANDSCAPE OF *MYCOSPHAERELLA GRAMINICOLA CYP51* UNDER SELECTION BY AZOLE FUNGICIDES

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The evolution of resistance to drugs and pesticides is a prime example of evolution in action. In many cases, such as the evolution of resistance to QoI and MBC fungicides in fungal pathogens of plants, the adaptive landscape is very simple, with a single mutation effectively conferring full resistance. However, in the case of the azole fungicides, multiple mutations in the target-site-encoding gene CYP51 occur in various combinations, with quantitative and epistatic effects on fungicide sensitivity and enzyme function. Epistatic interactions between mutations may result in rugged fitness landscapes, and incomplete cross-resistance between azoles causes the fitness landscape to shift as different fungicides are used. Mycosphaerella graminicola (Mycosphaerellaceae, Ascomycota) is a major foliar pathogen of wheat, causing yield losses of up to 50%. Azoles have been used extensively to control *M*. graminicola, but many CYP51 mutations have been reported, with up to eight coding changes in one haplotype, reducing the effectiveness of some compounds. Tools available to investigate *M*. *graminicola* CYP51 mutations include functional studies through heterologous expression and homologous gene replacement, experimental evolution through UV mutagenesis and *in vitro* selection, and historical studies with 160 years of archived samples from Rothamsted's Broadbalk long-term winter wheat field experiment. We consider the value of modelling the evolution of *M. graminicola CYP51* as an adaptive landscape, both in formulating anti-resistance strategies for azole use in agriculture, and in addressing fundamental evolutionary questions such as historical contingency and the crossing of adaptive valleys.

D23SY01RT11:42R2

SELECTION IN DIFFERENT LEVELS OF STRESS AND ITS EFFECTS ON FITNESS ACROSS A GRADIENT OF ENVIRONMENTAL CONDITIONS

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Most environments are variable over time or space, and the populations that are still surviving today have therefore most likely experienced many different environmental conditions. The performance of a population in a novel environment is likely to depend on its own particular selection history if the amplitude and direction of the correlation between the fitness landscapes of the past and novel environments is highly variable along a gradient of environmental conditions. We tested the prediction that adaptation to an intermediate level of stress would enhance performance across a gradient of permissive to stressful conditions by running a selection experiment in the unicellular alga *Chlamydomonas reinhardtii*. We selected replicated populations for about 40 generations in different levels of stress. We carried the experiment in six different environmental gradients and using three different strains to determine if the particular fitness landscape of the environment and/or the genetic architecture of the strain have an effect on the universality of the results. The results of this experiment will contribute in increasing our understanding of the effects of selection history on performance across a range of possible future environmental conditions.

D23SY01RT14:00R2

CAN WE PREDICT ADAPTATION TRAJECTORIES ON SIMPLE FITNESS LANDSCAPES?

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The dynamics of adaptation to a new environment is inherently complex, even in the simplest situations such as encountered in experimental evolution). Over the past decades, several empirical studies have measured the long-term dynamics of adaptation in different model species (mostly microorganisms). Yet, existing theory does not, to our knowledge, provide quantitative predictions to which such trajectories could be compared. Indeed, the speed with which a population adapts (i.e. the speed of the mean fitness increase) depends on the rate, fitness effects, and fate of beneficial mutations. While tremendous progress has been made in modelling these processes in steady state regimes, the key parameters are still difficult to measure, and more important, they vary over time in observed adaptive trajectories, because of background dependence effects (epistasis). The net result is that populations do not simply adapt linearly over time (i.e. at some steady state rate of increase), and the form of this nonlinearity is not predicted by any widely accepted model. Our goal here will be to show that some tools exist that provide testable predictions in this context, and check on a few examples if these predictions are accurate. We will first present some old and new results on Fisher's model and how they allow to predict the change in rate and effect of beneficial mutations over adaptive trajectories, from empirically measurable deleterious mutation effects and rate. Then we will discuss several alternative tools that can be used to model adaptation trajectories in these types of landscapes, accounting for non - stationary distributions of mutation effects and rates. We will illustrate the use of this approach on some empirical trajectories in model species.

D23SY01RT14:24R2

EXPERIMENTAL HOST-PARASITE COEVOLUTION IN TEMPORALLY VARIABLE ENVIRONMENTS

<u>Alison B Duncan</u>¹, Franck Jacob¹, Eike Dusi¹, Michael Hochberg¹, Oliver Kaltz¹ ¹Universite of Montpellier 2, France alison.duncan@univ-montp2.fr

Antagonistic, host-parasite coevolution is predicted to be an important driver of genetic change. Despite this, few examples exist from natural populations, possibly because temporal environmental variation may interfere with the co-evolutionary process. Here we investigate experimental coevolution between Pseudomonas fluorescens and its phage SBW2 Φ 2 in a fluctuating temperature environment, changing between permissive (28°C) and restrictive (32°C) conditions, for phage populations. We investigated how the frequency of environmental change influenced coevolution in populations experiencing temperature switches every 2, 4 or 8 days, over a 16-day period. Phage persistence is severely reduced at 32°C, thus we hypothesised that longer periods at 32°C would be detrimental for the phage. We found that coevolutionary dynamics under fine-grained fluctuations (every 2 or 4 days) did not differ from the constant 28°C control. Contrary to expectation, coarsegrained fluctuations (every 8 days) benefited phage populations, despite extended periods at 32°C. During periods of 32°C, bacteria flourished in an environment where phage were unable to infect, but also lost previously acquired resistance. Although infections did not occur at 32°C, all phage populations persisted. Accordingly, once populations were returned to 28°C, phage benefitted from high infection rates of susceptible bacteria. Our results highlight the need to consider temporal environmental heterogeneity when investigating coevolutionary interactions.

D23SY01RT14:48R2

GENETIC ROBUSTNESS AND ENVIRONMENTAL ROBUSTNESS IN AN RNA VIRUS

<u>Daniel Goldhill</u>¹, Paul E Turner¹ ¹*Ecology and Evolutionary Biology, Yale University, United States* daniel.goldhill@yale.edu

Robustness, the ability of phenotypes to withstand perturbation, is often categorized into environmental robustness, resistance to environmental change, and genetic robustness, resistance to mutation. Determining how robustness interacts with the fitness landscape is vital to increasing our understanding of how organisms successfully adapt to changing environments. Studies in proteins and RNA molecules have shown that areas of the fitness landscape with high environmental robustness have high genetic robustness, a property that has been termed plastogenetic congruence. However, it is unknown how environmental and genetic robustness relate at higher levels of biological organization such as organisms. We have experimentally evolved an RNA virus, $\phi 6$, to have higher resistance to heat shock, a form of environmental robustness. We have tested the virus for genetic robustness using a mutagen and shown that the evolved virus maintained higher fitness and therefore had higher genetic robustness. Previous work with this virus has shown that increased genetic robustness does not necessarily lead to increased environmental robustness. Our investigations of the mutations leading to increased robustness allow us to suggest a mechanism linking environmental and genetic robustness in an RNA virus. The study of robustness at the level of a virus provides insight into the nature of the fitness landscape and may help predict evolutionary trajectories.

D23SY01RT15:12R2

GENETIC INTERACTIONS BETWEEN BENEFICIAL MUTATIONS IN SACCHAROMYCES CEREVISIAE

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Independently evolving populations may adapt to similar selection pressures via different genetic changes. The interactions between such changes can then inform us about the underlying fitness landscape, allowing us to determine whether gene flow would be facilitated or hampered following secondary contact. We used Saccharomyces cerevisiae to measure the genetic interactions between independently evolved first-step mutations to the fungicide nystatin. We found that genetic interactions are prevalent, even among the first adaptive mutations. In the adaptive environment, the more beneficial mutation often masks the other, less beneficial one. This would allow a population fixed for the less beneficial allele to acquire and fix a more beneficial allele - thus continuing to climb the adaptive peak. In one case, however, reciprocal sign epistasis was observed, indicative of a fitness valley between two peaks. This is surprising given the small number of mutations combined and the relative simplicity of the adaptive environment.

POSTERS

EMPIRICAL FITNESS LANDSCAPES REVEALS A LIMITED NUMBER OF ACCESSIBLE ADAPTIVE PATHWAYS FOR AN RNA VIRUS

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RNA viruses are the main source of emerging infectious diseases owed to the evolutionary potential bestow by their fast replication, large population sizes and high mutation and recombination rates. However, an equally important parameter, which is usually neglected, is the topography of the fitness landscape, that is, how many fitness maxima exist and how well connected they are, which determines the number of accessible evolutionary pathways. To address this question, we have reconstructed a fitness landscape describing the adaptation of Tobacco etch potyvirus to a new host, Arabidopsis thaliana. Two fitness traits were measured for most of the genotypes in the landscape, infectivity and virus accumulation. We found prevailing epistatic effects between mutations in the early steps of adaptation, while independent effects became more common at latter stages. Results suggest that the landscape was rather smooth, with a high number of potential neutral paths and a single fitness peak.

D23SY01PS0107

EXPERIMENTAL EVOLUTION FOR GROWTH RATE AND ITS IMPLICATIONS FOR INFECTION SUCCESS, CO-INFECTION DYNAMICS, AND VIRULENCE IN A TRYPANOSOME PARASITE OF BUMBLEBEES

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Host-parasite interactions and their outcomes are strongly affected by several factors such as host and parasite genotypes and environmental conditions. Selection on basic growth properties in parasites may have far reaching consequences for numerous parasite traits, infection outcome and importantly the consequences for host fitness. It is know that strains of the trypanosome parasite of bumblebees *Crithidia bombi* have widely varying growth rates when cultured *in vitro*. We aim to experimentally evolve this parasite *in vitro* selecting for fast and slow growing sub-lines. This will enable us to investigate the costs, benefits and fitness trade-offs related to parasite growth rate by subsequently measuring *in vivo* infection profiles, transmission, and competitive ability under co-infection. To our knowledge, it would be the first time ecological trypanosome isolates have been experimentally evolved *in vitro*. These results will help define the fitness consequences for the observed natural variation in *C. bombi* growth, and will also inform important aspects of host-parasite evolution including the evolution of virulence.

VIRAL EXPERIMENTAL EVOLUTION REVEALS GENOTYPIC BUT NOT PHENOTYPIC HISTORICAL CONTINGENCY

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The importance of historical contingency in determining the evolvability of virus has been largely unappreciated. Identifying the constraints imposed by past adaptation is, however, of importance for understanding the evolution of host usage dynamics by multi-host viruses or the emergence of escape mutants that persist in the absence of antiviral treatments. To address this issue, we undertook an experimental evolution approach. Sixty lineages of *Tobacco etch potyvirus* that differ in their past evolutionary history and degree of adaptation to Nicotiana tabacum, and thus in their position on the adaptive landscape, were allowed to adapt to this host for 15 rounds of within host multiplication and transfer. The degree of adaptation to the new host as well as to the original ones was evaluated and the consensus sequence of each evolved lineage was obtained. Past evolutionary history did not determine the phenotypic outcome of this common host evolution phase. By contrast, historical contingency left footprints at the genotypic level: the majority of host-specific mutations present at the beginning of this experiment were retained in the end-point populations and may have affected which new mutations were consequently fixed. Further divergence between the sequences occurred despite a shared selective environment. Our results imply that viruses are not easily trapped into suboptimal phenotypes and that TEV (re)emergence is not evolutionarily constrained.

D23SY01PS0203

EXPERIMENTAL EVOLUTION UNCOVERS ENVIRONMENT DEPENDENT FITNESS DIFFERENCES AMONG WOLBACHIA STRAINS IN DROSOPHILA MELANOGASTER

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Natural populations of *Drosophila melanogaster* carry different *Wolbachia* strains, but it is not clear whether these *Wolbachia* strains are functionally diverged. To address this question, we exposed a natural *D. melanogaster* population to two different environments (hot and cold) and traced the relative frequency of different *Wolbachia* haplotypes during the experiment. Our *D. melanogaster* population sample from Portugal contained a highly polymorphic *Wolbachia* population in which three of the six described clades were present. Using Pool-Seq, we found that in the hot environment the *Wolbachia* composition remained remarkably stable over 37 generations. In the cold environment, however, *Wolbachia* strains from two clades increased in frequency. To distinguish between host and *Wolbachia* mediated effects, we exposed flies that evolved for 37 generations in the hot environment to the cold environment. Interestingly, we observed a very similar pattern to the previous experiment, suggesting that *Wolbachia* from two clades have a fitness advantage in the cold environment.

D23SY01PS0353

EVOLVING IN AN UNPREDICTABLE WORLD - THE E. COLI STORY

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Effects of temporally fluctuating environments on the fitness of populations are less explored compared to the effects of directional selection regimes. Existing studies on temporally fluctuating environments employ a narrow selection regime i.e. mostly one environmental parameter fluctuating predictably between two limit points and the fitness measurements happen in the environmental backgrounds similar to that of selection. Obviously such studies fail to predict the fitness outcomes in the complex and/or novel, unpredictable environments. We select replicate microbial populations under randomly fluctuating complex, stressful environmental regime. When fitness proxies of these selected lines are compared with the control populations grown in benign environments, under multiple novel environmental backgrounds, selected lines display 'Generalist' properties. We further characterize these Generalists by fitness proxy measurements at different time intervals during the growth in the novel environments. We see that acclimation is beneficial in both selected and control populations but consistently more advantageous in the populations with the history of randomly fluctuating environment. Characterization of the Generalists on the mechanistic level, shows that the commonly evoked explanations of evolution of hypermutators or modified permeability or carry over plasticity are not sufficient to explain observed Generalist phenomenon.

EXPERIMENTAL EVOLUTION OF HEAVY METAL TOLERANCE IN CHANGING ENVIRONMENTS

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In most long-term laboratory evolution experiments, organisms are exposed to a constant selection regime that initially causes a large reduction in fitness. However, the ecological relevance of this treatment may be questioned: under natural circumstances, environmental variables likely vary with time. We were interested in how the rate of directional environmental change affects the evolution of heavy metal tolerance in Saccharomyces cerevisiae. To this end, we grew replicate lines of yeast for 500 generations in the presence of (i) a constant high concentration of Cd, Ni or Zn or (ii) gradually increasing concentrations of these metals. We anticipated that these contrasting selection regimes would result in different adaptive dynamics and evolutionary endpoints, as the shape of the fitness landscape changes as a function of metal concentration. More specifically, we propose the following alternative scenarios: 1) the most resistant genotype is most fit at all metal concentrations, but strength of selection is proportional to concentration 2) the optimal genotype changes with concentration, such that the optimal genotype at intermediate concentrations will confer an intermediate level of tolerance. These scenarios predict that a gradual increase of metal concentration (as opposed to a constant high concentration) causes mutations of large effect to be fixed at later time points (scenario 1), only mutations of small or intermediate effect to be fixed (scenario 2) and, if the fitness landscape is rugged, evolutionary endpoints to be fitter and more diverse (both scenarios). Here, we present results from competition assays that were used to determine the relative fitness of evolved and ancestral isolates and thus differentiate between the alternative hypotheses. Although evolutionary dynamics differed between the treatments, evolutionary endpoints had a similar fitness, reflecting a smooth fitness landscape that changes as a function of metal concentration.

EVADING GENETIC DRIFT: AN EXPERIMENTAL TEST OF THE PROBABILITY OF FIXATION OF NEW GENETIC VARIANTS

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Progression through an adaptive fitness landscape requires the appearance and fixation of beneficial mutations. But fixation of a beneficial variant is not a necessary outcome, as deduced by J.B.S. Haldane in 1927, instead its probability can be given by twice the fitness effect: Pfix= 2s. This is based in the reasoning that even beneficial variants are lost with high probability by genetic drift. In this work, we performed invasion experiments with inbred lines of *Caenorhabditis elegans* in well-defined demographic conditions to experimentally demonstrate the determinant role of drift in the initial dynamics of new beneficial variants, as expected. We also show that the extinction of a deleterious variant, when at low frequency, is higher than that of a beneficial variant thus establishing that classical population genetics theory can accurately predict the fate of low frequency variants. Remarkably though we also find that, when at high frequency the fate of these variants is distinct from their low frequency dynamics, which results not in their ultimate fixation or loss, but on their maintenance. Our data confirm one of the key results of population genetics theory and highlights the complex nature of adaptation, where polymorphism can be maintained or lost depending on population structure.

D23SY01PS0539

THE EVOLUTION OF BACTERIAL MUTATION RATES UNDER SIMULTANEOUS SELECTION BY INTER-SPECIFIC AND SOCIAL PARASITISM

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Many bacterial populations harbour substantial numbers of hypermutable bacteria, in spite of hypermutation being associated with deleterious mutations. One reason for the persistence of hypermutators is the provision of novel mutations, enabling rapid adaptation to continually changing environments, such as coevolving virulent parasites. However, hypermutation also increases the rate at which intra-specific parasites (social cheats) are generated. Inter-specific and intra-specific parasitism are therefore likely to impose conflicting selection pressure on mutation rate. Here, we combine theory and experiments to investigate how simultaneous selection from inter- and intra-specific parasitism affects the evolution of bacterial mutation rates in the plant-colonizing bacterium *Pseudomonas fluorescens*. Both our theoretical and experimental results suggest that phage presence increases, and selection for mutator bacteria. Moreover, phages imposed a much greater growth cost than social cheating, and when both selection pressures were imposed simultaneously, selection for cooperation did not affect mutation rate evolution. Given the ubiquity of infectious phages in the natural environment and clinical infections, our results suggest that phage are likely to be more important than social interactions in determining mutation rate evolution.

REPRODUCTIVE ASSURANCE DRIVES MATING SYSTEM TRANSITIONS TO SELFING

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Evolutionary transitions from outcrossing to selfing are thought to occur because selfing reproductively assures the persistence of populations when mates or pollinators are unreliable. Direct evidence for reproductive assurance is however equivocal, in particular because outcrossing can easily overcome the adaptive benefits of selfing. To test for the impact of reproductive assurance by selfing, we performed invasion experiments of hermaphrodites into predominantly male-female populations of the nematode Caenorhabditis elegans. The invasion and fixation of selfing resulted in adaptation when populations evolved in a new environment that hindered mating. In contrast, when selfing failed to invade, adaptation to the novel environment was compromised because mating was possible. Using Monte Carlo simulations we further explored the factors that might have influenced the invasion dynamics of selfing. We confirm that reproductive assurance was responsible for the experimental transition from outcrossing to selfing.

EXPERIMENTAL EVOLUTION OF AN RNA VIRUS IN MIXED HOST ENVIRONMENTS

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Spatially heterogeneous environments pose unique challenges for evolving asexual populations. One possibility is that selection will lead to high genetic variance, where multiple subpopulations adapt to specialize on a subset of the available habitats. In contrast, spatial heterogeneity may lead to evolution of generalism, where the population is dominated by a single genotype with a broad niche. However, the evolution of populations in spatially heterogeneous environments has been rarely examined outside of theory, and to date there have been no experiments that test how rapidly evolving RNA virus populations adapt in the face of this challenge. Here, we allowed vesicular stomatitis virus (VSV) to evolve in replicated environments containing different mixtures of two host types in laboratory tissue culture: cancer-derived HeLa cells and non-cancerous BHK cells. After 25 passages (100 generations), the fitness (growth) of evolved viruses was assayed on each host type, relative to the common ancestor. We observed a correlated response to selection when VSV was evolved on pure cultures of either host (100% BHK or 100% HeLa). However, in mixed environments this correlated response was broken; populations evolved in spatially heterogeneous environments consistently improved on HeLa cells, even when HeLa cells were rare, but were much more variable in their improvement on BHK cells. Current work examines whole-genomics of evolved populations, to determine how simple versus spatially-complex environments affect genetic variation in VSV populations, and to compare/contrast trajectories of phenotypic and genetic change within and among treatments. This study demonstrates that simple versus mixed environments can pose fundamentally different challenges for adapting populations, and the need for theory that addresses how environmental complexity may influence adaptive trajectories.

EXPLORING THE VALIDITY OF REPRESENTING HIGH-DIMENSIONAL FITNESS LANDSCAPES WITH A QUADRATIC APPROXIMATION

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A predictive model for in vitro replicative fitness of HIV, that estimates main fitness effects and epistatic interactions of mutations with a generalized kernel ridge regression, was presented in [1]. The predictive power of the resulting quadratic approximation was found to be high when testing on an independent dataset. A quantitative analysis of the resulting fitness landscape further showed that it was less rugged, less neutral and more correlated than expected [2]. However, due to the highdimensionality of the sequence space it is intractable to assess its performance on all possible sequence variants. We evaluate the regressor's generalization performance by approximating quasi-empirical fitness landscapes based on RNA secondary structure. This is the only known system where a genotype-phenotype map can be easily and accurately computed [3]. Although fitness does not solely depend on RNA secondary structure, it is a good proxy. Unbiased datasets and datasets biased to contain sequences close to the target genotype or phenotype are simulated. Fitness is measured as a combination of thermodynamic stability and the distance to a target structure. The model is alternately trained and tested on each pair of datasets. In addition, the approximations are compared to the real quasi-empirical fitness landscapes with respect to ruggedness, neutrality and correlation length. Together, this provides an analysis of the validity of using a quadratic approximation to represent highdimensional fitness landscapes. [1] Hinkley, T., Martins, J., Chappey C. et al. A systems analysis of mutational effects in HIV-1 protease and reverse transcriptase. Nat Genet, 43, 2011. [2] Kouvos, R.D., Leventhal, G.E., Hinkley, T. et al. Exploring the complexity of the HIV-1 fitness landscape. PLoS Genet, 8(3) 2012. [3] Schuster, P., Fontana, W., Stadler, P.F. and Hofacker, I.L., From sequences to shapes and back: A case study in RNA secondary structures. Proc R Soc B, 255(1344) 1994.

D23SY01PS1054

EVOLUTIONARY CONSEQUENCES OF FEMALE PROMISCUITY

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To cope with stressful conditions like parasite attacks or temperature changes, females can increase their offspring diversity and fitness by mating multiply with several males. This can result in male harm but can be advantageous in fluctuating environments. Populations with promiscuous females therefore are supposed to be more stable than populations with monogamous females. We study the gonochoristic nematode *Caenorhabditis remanei*, which is mating multiply and affected by male harm. Using experimental evolution, we were able to show that females, in contrast to males, were able to adapt to sex ratio manipulations. Additionally, offspring of promiscuous females had a higher fitness than monogamous females under the influence of the microparasite *Bacillus thuringiensis*. Even though a powerful tool to investigate fitness effects, manipulating sex ratios in nematodes in scientifically meaningful numbers is very work intensive if done by hand. To increase the population size and replicate number and therefore decrease the effects of drift, we are using differently labeled males and females and an automated sorting device. Worms are loaded onto a polymeric silicon chip (PDMS) and screened for fluorescent makers, resulting in separated males and females, which can be combined in different sex ratios depending on the treatment. These populations will be tested under stressful conditions to investigate whether populations with promiscuous females are more resistant to stressful changes in their environment compared to the monogamous control. Additionally, individual females will be compared to analyze within and between population fitness.

1. Experimental Evolution on Empirical Fitness Landscapes

D23SY01PS1186

SOCIAL INTERACTIONS DETERMINE MUTATION RATE IN ESCHERICHIA COLI

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Spontaneous mutation is the key engine of evolution. It is fully appreciated that mutation rates vary between genotypes and locally over the genome, but how plastic mutation rate can be for a particular site in a particular genotype is far less clear. I will describe our recent findings that rate of mutations conferring rifampicin resistance increase with decreasing density of *Escherichia coli* cells, and that this mutation rate plasticity is mediated by genes involved in quorum sensing. I will present a view that mutation rate is a plastic trait that can be regulated by cell-cell communication, and go on to discuss how our results might help us to better understand the emergence of *de novo* genetic resistance in microbial communities.

1. Experimental Evolution on Empirical Fitness Landscapes

D23SY01PS1391

SHORT-TIME RESPONSE FUNCTION OF THE MODULAR DILUTE SHERRINGTON-KIRKPATRICK MODEL

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We consider the short-time energy relaxation of the dilute SK model. We show that the more modular the system, the more rapidly the energy decays at short times. Conversely, a more modular system reaches a less favorable energy at long times in a static environment. We use these results to discuss the dynamics of the modularity order parameter in a system for which the coupling parameters of the dilute SK model change in time, due to a changing environment. Modularity endows the spin glass with a better response function in a changing environment.

Symposium

2. Selection and Evolution in Natural Populations

22, 23 and 24 August



Program

Thursday 22 August

Session(s): 9

Friday 23 August

Session(s): 10, 11, 12, 13

Saturday 24 August

Session(s): 14

Organisers: Tom Tregenza and Trine Bilde

Invited speakers: Ary Hoffmann, Susan Johnston and Jane Reid

Description:

This symposium will showcase studies designed to understand evolution by monitoring natural populations over multiple generations. It will highlight new model systems as well as recent discoveries in established systems. There will be a particular emphasis on innovative approaches to measuring selection in the wild, including new methodologies for measuring the success of genes and individuals. These will include the use of genetic markers, exploitation of natural pedigrees and new tagging and monitoring approaches.

D22SY02IT10:30R1

GENE MAPPING REVEALS WHY SEXUALLY-SELECTED GENETIC VARIATION IS MAINTAINED IN A WILD MAMMAL POPULATION.

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Sexual selection is assumed to be a source of strong and sustained directional selection in the wild, and is expected to lead to a decrease in underlying genetic variation. However, there is often considerable genetic variation in sexually-selected traits in wild populations and consequently, this phenomenon has become a long-discussed paradox in the field of evolutionary biology. In wild Soay sheep, large horns confer an advantage in strong intra-sexual competition, yet both sexes have an inherited polymorphism for horn type and have substantial genetic variation in their horn size. We pinpointed the causal genetic mechanisms influencing horn development using genome-wide association studies (GWAS) and fine-scale SNP genotyping. This allowed us to test the most commonly proposed explanations for the maintenance of sexually-selected genetic variation, such as genic capture ("good genes"), sexually antagonistic selection, and trade-offs between sexual and non-sexual fitness (i.e. reproductive success vs. survival). I present the evidence for and against these mechanisms in the Soay sheep population, and discuss the applications of these findings to other wild populations.

D23SY02IT10:30R1

CLINAL SELECTION IN DROSOPHILA: WHAT HAVE WE LEARNT, WHERE ARE WE GOING?

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Clines have traditionally been used to investigate patterns of selection on phenotypes and genotypes involved in climatic adaptation. More recently, clines are being used to test for changes in patterns of adaptation across time. Modern genomic and transcriptomic approaches are now being applied to understand patterns of clinal adaptation at a fine level and to allow new sets of questions to be considered and a new level of understanding to be reached. Here I focus on recent Drosophila work to overview the opportunities and challenges provided by these new approaches and how they can be integrated into the more traditional work. I also consider the ways in which experimental work in field and laboratory settings can be used to further understand patterns of selection.

D24SY02IT10:30R3

QUANTITATIVE GENETIC COVARIANCES UNDERLYING SELECTION ON POLYANDRY IN A NATURAL SONG SPARROW POPULATION

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Understanding the forces that drive the evolution and persistence of female polyandry remains an overarching yet elusive goal in evolutionary ecology. Many key hypotheses explaining selection on polyandry are explicitly quantitative genetic, involving genetic covariances between polyandry and fitness components in females and their offspring, and cross-sex genetic covariances with components of male fitness. However, despite recent methodological advances, these hypotheses have not previously been explicitly tested by estimating key genetic covariances in wild populations experiencing natural variation in female polyandry, male mating success and associated fitness components. I apply quantitative genetic analyses to 20 years of comprehensive paternity and pedigree data from polygynandrous song sparrows (Melospiza melodia) to test the key hypotheses that 1) there is additive genetic variance in the degrees of female polyandry and male paternity success, 2) that polyandrous females produce offspring of higher additive genetic value through extra-pair reproduction, and 3) female polyandry is positively genetically correlated with male paternity success. I demonstrate substantial additive genetic variance in both female polyandry and male paternity success, potentially allowing rapid evolution in both traits. I show that, opposite to prediction, polyandrous females produce offspring of lower additive genetic value through extra-pair reproduction, implying a component of indirect selection against polyandry. Finally, I demonstrate positive genetic covariance between female polyandry and male paternity success, but show that this covariance is constrained by genetic covariances among male fitness components. Through this explicit quantitative genetic approach I identify components of selection that could explain the evolution of polyandry and the current maintenance of genetic variation in polyandry under conflicting components of sex-specific selection.

D22SY02RT11:18R1

AGE RELATED CHANGES IN FITNESS TRAITS IN A WILD FIELD CRICKET POPULATION

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Evidence for senescence in nature has been found in a growing number of vertebrate species, particularly birds and mammals, and more recently in a few invertebrates. Insects have proved very suitable for the study of senescence in laboratory studies but their mobility and small size make them very difficult to follow in their natural environment. We have extended the study of insect ageing into the wild, by using up to 90 video cameras to continuously monitor a natural field cricket population. We tagged individuals immediately after they became adult and followed them until their death was either inferred or directly observed. We recorded mating and fighting success, singing activity and movement over the entire adult life of individual crickets for five consecutive generations. We examine changes in behaviour over the adult lifespan and relationships with reproductive success. This allows us to determine whether crickets senesce in our population in terms of both survival probability and changes in behavioural measures such as activity, and to ask questions about interactions between ageing and sexual selection. Our model system provides an excellent opportunity for the study of the selective pressures acting on life history and ageing in wild insects.

D23SY02RT19:16R1

REJECTION OF THE CLONAL EROSION HYPOTHESIS: A STUDY OF HYBRIDIZING DAPHNIA FROM TEN NEIGHBORING LAKES

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Cyclical parthenogenetic water fleas of the genus *Daphnia* are commonly used as a model system in ecological and evolutionary research. On the one hand, their capability of sexual reproduction favours genetic variation not only within species, but also between species via interspecific hybridization; on the other hand, asexual reproduction may lead to clonal erosion. In the present study, we tracked ten communities of the *D. longispina* complex (maximum distance about 30 km) from the beginning to the end of one growing season, using 15 microsatellite loci. The clonal richness remained roughly constant throughout the growing season and all four lakes, leading us to reject the clonal erosion hypothesis. Moreover, some identical multilocus genotypes from parental species were shared among otherwise unconnected lakes, and the genetic setup of the communities and populations matched well with the geographical positions among the lakes; but only for parental species and not for the hybrids. This indicates either strong contemporary gene flow or past colonisation events from neighbouring populations. Overall, the genetic distance among populations was lowest for *D. galeata*, confirming its invasive nature, modest for *D. longispina* and the highest for hybrids. Thus, hybrids appear to be locally produced, rather than migrating from neighbouring lakes.

D23SY02RT11:18R1

POLLINATOR-MEDIATED SELECTION ON FLORAL SIZE AND COLOR IN TWO IRIS SPECIES

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Pollinator-mediated selection is considered the major drive of floral evolu-tion and the evolution of floral diversity among angiosperms. Yet, causes of selection on traits contributing to attract pollinators have rarely been tested experimentally in natural populations. We quantified phenotypic selection on floral size and color in two species of the Oncocyclus irises in two years, and experimentally assessed the contribution of pollinators for this selection. Flowers of the Oncocyclus irises are among the largest in the East Mediter-ranean flora, and are usually dark-colored, from purple to black. Floral color is utilized to absorb the morning sun rays and to increase temperature within the flower. This morning floral heat rewards the pollinators of the irises: male solitary bees that shelter overnight in the flowers. To determine whether selection on floral size and color can be attributed to interactions with pollinators, selection was quantified for both open-pollinated controls and for plants receiving supplemental hand-pollination. The latter treatment is expected to remove pollinator limitation and hence to relax pollinator-mediated selection. Supplemental hand-pollination changed the extent and direction of selec-tion, suggesting pollinator-mediated selection on these traits. Pollinator-mediated directional selection on floral size and color was detected in Iris atropurpurea, and non-linear pollinator-mediated selection was detected in Iris haynei. This study provides evidence that pollinators of these irises are the selec-tion agent on floral traits, and support the contention that pollinators can drive the evolution of floral display size and color.

D23SY02RT11:42R1

EVOLUTIONARY POTENTIAL OF ARRIVAL DATE ON BREEDING GROUNDS: SELECTION, HERITABILITY AND MICROEVOLUTION

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In migratory species, timing of arrival on the breeding grounds in spring is crucial for the reproductive success. Thus, in the face of a changing environment, it is important to be able to adjust the timing accordingly. However, little is known about the genetic background and hence evolutionary potential of arrival date. We have used a multi-level approach to investigate the evolutionary potential of arrival date combining data from a multigenerational pedigree and novel migration tracking techniques of a natural population of great reed warblers (*Acrocephalus arundinaceus*). We found that selection favours early arrival both in male and female great reed warblers, and that the trait is both repeatable and heritable. Further, arrival date in the population has advanced during the two decades of this study, a pattern that is in accordance with the response attributed to climate change reported in other migrant birds but also in accordance to the directional selection acting on the trait. Tracking the full migratory annual cycle of individual great reed warblers show that departure date from the wintering site determines arrival date and that spring migration is faster than autumn migration, corroborating the selection for earlier arrival. Our study is a first step towards dissecting the genetic and environmental factors that contribute to shape arrival date in long-distance migrant birds. Such analyses are essential if we want to understand how migratory species are able to cope with a rapidly changing environment.

D23SY02RT14:00R1

EVOLUTIONARY CHANGES IN THE SEXUAL REPRODUCTIVE SYSTEM: THE CASE STUDY OF THE INVASIVE TRISTYLOUS OXALIS PES-CAPRAE

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The establishment and spread of invasive species depends on different evolutionary and ecological pressures, with reproduction being a key factor for success. Among other strategies, vegetative propagation has been correlated with invasion potential. Because clonality affects the spatial distribution of genets and its flowers, clonal species are expected to have increased rates of selfpollination. However, selfing might be advantageous as it enables to overcome the lack of compatible mates or scarcity of pollinators during invasion. Under this scenario, in self-incompatible species, selection may favor the breakdown of the incompatibility system, as plants with some levels of compatibility would have advantage in low density conditions and be able to establish new populations after dispersal (Baker's law). Oxalis pes-caprae is a tristylous species with heteromorphic incompatibility; in most invaded areas, strong founder events lead to the introduction of the 5x shortstyled morph only, leading to an exclusively asexual mode of reproduction. Yet, recent studies have shown scattered sexual reproduction and the emergence of new floral morphs in the western Mediterranean basin. Our objective was to evaluate the rates of self- and morph-incompatibility across this invaded area, and compare it with the native range, in order to assess changes in the reproductive system. Controlled hand-pollinations were made in invasive and native populations and plant reproductive success was quantified. The incompatibility system varied among invasive populations and floral morphs; overall a breakdown in several populations across the invaded range was observed, mostly through a partial breakdown in morph-incompatibility of S-morph, but also in selfincompatibility system of M- and L-morphs. Because reproductive strategies determine the demography and genetics of invasive populations, the results obtained are important to understand the evolutionary dynamics operating during invasion.

D23SY02RT14:24R1

RECOMBINATION LANDSCAPE OF FICEDULA FLYCATCHERS

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Characterizing variation in recombination rates between and within species is essential for understanding the patterns of gene flow, the efficacy of selection, and the variation in genetic divergence and diversity. The Natural pedigree of collared flycatchers (*Ficedula albicollis*) available from the Baltic island of Öland provides an exclusive opportunity to investigate variation in the rate of recombination in wild bird populations. Using a newly developed custom 50,000 SNP chip, we constructed a high-density genetic linkage map of the collared flycatcher. Our linkage analysis placed 4,951 SNPs in 33 linkage groups, corresponding to 29 autosomes and the Z chromosome. The total genetic distance was 3,096 cM with higher recombination rate in males than females (3,226 cM in males, 2,970 cM in females). Recombination rates substantially vary between chromosomes (mean recombination rate per chromosome = $2.04 \sim 6.97$ cM/Mb) as well as within chromosomes and are generally higher toward the end of chromosomes. In addition to the collared flycatcher pedigree samples, we have genotyped multiple populations of collared flycatchers across Europe to characterize the pattern of linkage disequilibrium (LD) and estimate population-scaled recombination rate (rho). Population-specific recombination patterns and association with underlying molecular features will be discussed.

D23SY02RT14:48R1

HOW SOCIAL STRUCTURE AFFECTS GENE FLOW IN A WILD PASSERINE POPULATION

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Gene flow is strongly affected by the spatial distribution of individuals, the variability of the physical environment and social processes such as dispersal, resource competition and territoriality. Here we investigate the contributions of those forces to gene flow in foraging flocks of great tits (*Parus major*) for three consecutive winters. We used a total of 85602 visits of flocks to 60 feeding tables which recorded the identity of 1711 birds by radio frequency identification technology. Of those birds 962 were genotyped based on 4701 autosomal single-nucleotide polymorphisms (SNPs). For 87% of the visits we were able to genotype at least one individual. We used asymmetric eigenvector maps (AEMs) to partition the contributions of space and social structure to the allele frequencies of all 4701 SNPS in the feeding flocks while taking the previous positions of individuals into account. We were able to explain 58% of the variance in allele frequencies with AEMs. This study shows that space and social structure have a substantial effect on the distribution of alleles over subpopulations and therefore on gene flow. To our knowledge this is the first study to investigate the effect of social structure and space to gene flow at such a fine scale while accounting for previous location of the individuals. Therefore it contributes to the understanding of how social behaviour affects evolution. Next we will extract Moran's eigenvector maps (MEMs) for the spatial locations of feeding tables and use partial redundancy analysis (AEMs as explanatory variables while controlling for MEMs) to investigate how social structure affects allele frequencies, while controlling for space. We will use variance partitioning to quantify the relative contributions of space and social structure, determine which alleles have large effects on the AEMs and check whether those alleles are in linkage disequilibrium with candidate genes or are known to correlate to environmental variables.

D23SY02RT15:12R1

INDIRECT GENETIC AND ENVIRONMENTAL EFFECTS ON PARENTAL AND ALLOPARENTAL EFFORT

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The partition of phenotypes into genetic and environment effects can be expanded by considering that an individual's environment is also made up of other individuals. In social contexts, then, the phenotype of a target individual can be affected by the phenotypes and, therefore, the genotypes, of others it interacts with, and vice versa. These indirect genetic effects are an additional source of genetic variance with which selection can act. We used a cooperative breeding species of birds, long-tailed tits, as a model system to study social effects among family groups. This species suffers from high rates of nest predation and individuals who fail to breed in a given year become helpers at the nest of a close relative. Being helped is linked to both offspring and breeder survival. We investigated indirect genetic and environmental effects on provisioning effort among members of a breed group. We found a negative correlation for feeding rate between the within-year environment effects that an individual has on themselves and on the other members of their breeding group. However, these indirect effects were not heritable and had low repeatability across years. Birds thus differ in their ability as helpers and this has consequences for the total performance of the group beyond their individual contribution because the members of the breed group adjust their effort in response to each other. The lack of substantial genetic and permanent environment variation indicates that effort is largely a matter of yearly condition and suggests that provisioning effort is facultative to optimize fitness through both offspring recruitment and individual survival.

D23SY02RT15:45R3

SELECTION ON POST-REPRODUCTIVE LIFESPAN IN ASIAN ELEPHANTS

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Among terrestrial mammals, elephants share the features with great apes and humans, of having long lifespan and offspring with long dependency. In humans, these traits combine with female menopause and an extended post-reproductive lifespan. Some have suggested that elephants, too, have a comparable post-reproductive lifespan to women, with survival into 60s in the wild and max. known age >80yrs. However, little data exists on the frequency of post-reproductive lifespan or its fitness benefits in long-lived species except humans. Long lifespan after last birth has been proposed to evolve because of the long offspring dependency and detrimental effects of early mother loss on their fitness. Here we use extensive (n>8000) individual-based multigenerational demographic records on semicaptive Asian elephants in Myanmar to investigate first, the patterns of post-reproductive survival in females and second, the short- and long-term importance of maternal care to offspring survival. We found that first, the age-specific fecundity clearly decreases after age 50, but the pattern does not correspond to the total loss of fecundity in old age found in human females. The elephant postreproductive phase covers only one tenth of the whole lifespan, in contrast to almost half of that in women. Secondly, maternal death during the first years reduces calf survival considerably, but such effects wane rapidly with age, so that beyond age 5, mother's death no longer increases calf risk of death. Calves surviving mother's immediate death do not show long-term effects in later ages. Thus, Asian elephants show decreased fertility in advanced ages, but it differs distinctly from human agespecific fertility and the subsequent post-reproductive survival pattern. Our results imply that the long lifespan in elephants is not sufficiently explained only by long needed maternal care for calves and more generally, that longevity by itself does not necessarily lead to evolution of menopause.

D23SY02RT16:09R3

EVIDENCE FOR COST OF SEX AND PARASITE-MEDIATED SELECTION IN A NATURAL POPULATION OF CO-EXISTING SEXUAL AND ASEXUAL SNAILS

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One of the main hypotheses for why sexual reproduction is so common despite the cost of producing males proposes that negative frequency-dependence selection by co-evolving parasites provides an advantage for rare genotypes. However, this mechanism fails to predict advantage to sex when asexual assemblages are diverse. The challenge to sex posed by a diverse set of asexual lineages depends on the extent to which they realize their theorized two-fold advantage. Here, we used field-based comparisons of the rate of population growth between obligate sexual vs. multiple lineages of obligate asexual Potamopyrgus antipodarum, a New Zealand freshwater snail. We also used a population genetics approach to evaluate whether the temporal changes of population genetic structure follow patterns expected under parasite-mediated selection: faster clonal turnover in locations where parasite pressure is higher. The reproductive output of asexual lineages measured using experimental enclosures (cages anchored to the bottom of the lake) over a course of one year was as high as that of the best sexual families, which implies the cost of sex. We also found that the reproductive output of asexual lineages depends on the habitat, which implies that environmental heterogeneity may select for habitat-specific clonal assemblages. The genetic structure of the asexual population changed significantly over a 4-year period (4-8 generations) in shallow and mid-water habitats (high parasite pressure), but not in the deep habitat (low parasite pressure).
Our results show that the fitness of many asexual lineages is high enough to impose the cost of sex and that clonal turnover is faster in high infection sites. While the latter result is consistent with the parasite hypothesis for the maintenance of sex, the high relative fitness of many asexual lineages suggests that other mechanisms are needed to explain the persistence of sex in the face of a diverse array of asexual competitors.

D23SY02RT16:33R3

GENETIC AND ECOLOGICAL CONSEQUENCES OF INDIVIDUAL GENERALIZATION IN PLANT-POLLINATOR SYSTEMS

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Most theoretical approaches assume that, in pollination generalist systems, all individual plants interact with random subsets of the overall pollinator pool. In contrast, we think that the generalization degree of a given individual plant could be non random, but related with some intrinsic and extrinsic factors, such as its spatial location within the population, its microenvironment and its phenotype and genotype. Plants exhibiting different values for those factors would attract different subset of pollinators, and plants showing similar values of those traits would share similar subgroups of pollinators. Since pollinator species of a generalist plant vary in their foraging behaviour, per-visit effectiveness, and floral constancy, individual plants could receive a biased sample of pollen that could affect their reproductive success. To test this idea, we have thoroughly sampled a population of *Erysimum* mediohispanicum (Brassicaceae) in Sierra Nevada Mountains (Spain). This species is an extremely generalist plant, being pollinated by more than 180 insect species in the study area, ranging from 30-40 pollinator species per population. For every plant in the population, we determined the diversity and identity of insects visiting its flowers. In addition, we also quantified the micro-environment (irradiance, soil water content, top soil nutrients), the pollination-related phenotypic traits (flower number, flowering stalk height, stigma exertion, and corolla size, color, and shape), and the multilocus genotype (using cpDNA haplotypes and nuclear microsatellites) of each plant. Finally, to estimate individual fitness, we quantified the number and genetic diversity of the seeds produced per plant. Our goal in this study is to evaluate how the individual interaction with subsets of pollinators may affect the quantity and quality of plants' progeny.

D23SY02RT16:57R3

MUTUALISTS AND ANTAGONISTS DRIVE AMONG-POPULATION VARIATION IN SELECTION AND EVOLUTION OF FLORAL DISPLAY IN A PERENNIAL HERB

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Spatial variation in the direction of selection drives the evolution of adaptive differentiation. Yet, few experimental studies have examined the relative importance of different environmental factors for variation in selection and evolutionary trajectories in natural populations. We combined 8 years of observational data and field experiments to assess the relative importance of mutualistic and antagonistic interactions for spatial variation in selection and short-term evolution of a genetically based floral display dimorphism in the short-lived perennial herb Primula farinosa. Natural populations of this species include two floral morphs: long-scaped plants that present their flowers well above the ground and short-scaped plants with flowers positioned close to the ground. The direction and magnitude of selection on scape morph varied among populations, and so did the frequency of the short morph (median 19%, range 0-100%; N = 69 populations). A field experiment replicated at four sites demonstrated that variation in the strength of interactions with grazers and pollinators were responsible for among-population differences in relative fitness of the two morphs. Selection exerted by grazers favored the short-scaped morph, whereas pollinator-mediated selection favored the long-scaped morph. Moreover, variation in selection among natural populations was associated with differences in morph frequency change, and the experimental removal of grazers at nine sites significantly reduced the frequency of the short-scaped morph over eight years. The results demonstrate that spatial variation in intensity of grazing and pollination produces a selection mosaic, and that changes in biotic interactions may trigger rapid genetic changes in natural plant populations.

D23SY02RT17:45R3

UNCOVERING ADAPTATIVE DIFFERENTIATION IT THE WILD

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Differentiating in between adaptive and non adaptive causes of population differentiation in phenotypic traits of ecological interest remains often a challenge for empirical studies of natural populations. I will review a series of yet unpublished empirical studies which have utilized newly developed quantitative genetic approach to differentiate in between natural selection and genetic drift as a cause of population differentiation. Apart from of aiming to illustrate the general utility of this approach, I hope introduce couple of illuminating empirical case-studies where footprints of adaptive differentiation can be recovered in spite of strong influence of genetic drift and gene flow, respectively.

D23SY02RT18:09R3

INTERACTIVE EFFECTS OF PATERNAL GENOTYPE AND GROWTH ENVIRONMENT ON OFFSPRING CONDITION: A CROSS-FOSTERING EXPERIMENT IN THE PIED FLYCATCHER

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Conspicuous secondary sexual characters, such as sexually dichromatic plumage traits, are thought to have evolved through sexual selection, because they indicate the genetic or phenotypic quality of the bearer. While directional selection should deplete genetic variation in fitness-related traits, there are many cases where variation is higher than expected. One possible explanation for this is that different phenotypes within a population are adapted to different environmental conditions. The dorsal plumage coloration of pied flycatcher (*Ficedula hypoleuca*) males varies from almost completely black to dull brown. In addition they have several ornamental patches that also vary greatly in size and shape. We compared the condition and survival of the offspring of male pied flycatchers with different plumage phenotypes under different conditions. In order to create different environmental conditions, we used a partial cross-foster design where the original brood size was pairwise reduced or enlarged by one chick. This design enabled us to separate the effects of offspring genotype from parental effects. Preliminary analysis of the data indicates that there are significant interactions between the phenotype of the biological father (forehead patch size) and environmental conditions (brood manipulation treatment and ambient temperature) on chick condition. This suggests that offspring of different male phenotypes differ in their responses to environmental conditions. Therefore, selection on the trait is likely to be context-dependent and environmental heterogeneity may act as a factor maintaining phenotypic variation.

D23SY02RT18:33R3

SEX-BIASED DISPERSAL, INBREEDING AND FITNESS IN A SOCIAL INSECT

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Sex-biased dispersal may act as inbreeding avoidance strategy, yet data on natal dispersal and its effectiveness in reducing inbreeding in the wild are scarce. Perennial ant colonies offer a unique system to investigate the relationship between natal dispersal and inbreeding avoidance, as the dispersal and mating behavior of a queen during the short period of a mating flight will determine the genetic diversity among her offspring, for the entire life span of the incipient colony. We used pedigree information on parents of incipient and established colonies of the ant Formica exsecta, in order to examine the relationship between inbreeding, fitness, mating behavior, sex ratios, and patterns of natal dispersal. Dispersal was male-biased with male dispersal distances (ca. 150m) twice the length of those of queens (ca 60m), yet inbreeding was rife in the population. Neither multiple mating nor increasing dispersal distance by the queen reduced inbreeding among the offspring of her future colony. Queen homozygosity did not affect dispersal, but more homozygous queens had lower colony founding success, and were more incestuously mated themselves which may accentuate the negative effects of inbreeding. Inbreeding also affected resource allocation to the sexes, with inbred colonies producing smaller males, but not queens. We present first true estimates of natal dispersal distance in social insects. Our results emphasize that inbreeding may persist in absence of sib-mating, in species with supposedly population-wide panmictic mating behavior and sex-biased dispersal.

D23SY02RT18:57R3

ONTOGENETIC EFFECTS CAN INFLATE ESTIMATES OF GENETIC VARIANCE DEPENDING ON PEDIGREE STRUCTURE

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For reliably predicting micro-evolution it is crucial to properly estimate the trait's heritability. In natural populations heritabilities are often estimated using 'animal model' analyses that are based on pedigrees. As these data from natural populations are observational it leaves the possibility that the resemblance between related individuals is not due to shared genes but, e.g., to ontogenetic effects. Such effects could possibly severely bias heritability estimates of breeding time in birds: when the environment for the offspring provided by early breeding pairs differs from that by late breeding pairs and the breeding time of these offspring when they reproduce themselves is affected by this environment, this may lead to inflated heritability estimates. Using simulation studies based on data from two long-term study population of great tits (*Parus major*) we tested whether and how much such an early environmental effect can inflate heritability estimates from 'animal models'. We showed that early environmental effects can inflate heritability estimates depending on pedigree structure. We also used data from a wild population of great tits to directly test potential early environmental effects by comparing breeding time of females born early in the season in first broods and with that of sisters born much later in the season, namely in second broods. These birds are raised under very different environmental conditions but have the same genetic background. The egg-laying dates of first and second brood offspring do not differ when they reproduce themselves, clearly showing that ontogenetic effects are very small and hence family resemblance in timing is due to genes. Our results hence show that while ontogenetic effects do not affect an individual's breeding time as an adult, similar early environmental effects could potentially inflate quantitative genetic parameters from 'animal model' analyses depending on pedigree structure.

D24SY02RT11:18R3

ADAPTIVE DIVERGENCE IN TWO COEXISTING STICKLEBACK SPECIES

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Natural populations are at least partially adapted to local environmental conditions. The level of adaptation is promoted by divergent selection on ecologically relevant traits, but hampered by gene flow. The relative strength of both forces depends on genomic architecture as well as dispersal abilities, both of which might differ between species. In this study, we tested for divergent selection and gene flow in two coexisting and phylogenetically related species, the three- and nine-spined stickleback. Populations occur across very similar environmental clines in biotic and abiotic conditions (salinity, oxygen concentration, predation, parasites and prey), and were sampled within exactly the same spatial matrix. In each species we assessed gene flow between populations from contrasting environments and determined divergence in ecologically relevant traits (body shape, armour, spleen size, and trophic morphology). An analyses of genome-wide differentiation was used to identify the genomic basis of adaptation. Based on these data we compared the relative strength of divergent selection and gene flow in both species, as well as the biological functions of the genes under selection. We discuss how these relationships explain how both stickleback species maintain partially overlapping habitats.

D24SY02RT11:42R3

DRAWING A NATURAL FITNESS LANDSCAPE IN SPACE AND TIME: TEMPORAL ADAPTATION OF SOIL BACTERIAL COMMUNITIES

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Microbial communities are fundamental for ecosystem function, yet only little is known about their in situ ecology and evolution. How are such communities structured in space and time and which environmental factors drive their adaptation? We obtained soil bacterial isolates from three sites in a spatially structured design every month for eight months, representing a complete growth season. In a fully factorial transplant experiment we measured fitness of all isolates in media mirroring the environmental conditions in the soil sample they were isolated from (their "home" soil) and compared it to their growth rates in media representing soil at different spatial and temporal distances, thus not only describing the spatial fitness landscape for each isolate but also how the shape of this landscape changes over time. In comparison to growth under "home" soil conditions, growth rates were steadily declining in media representing future soil conditions, indicating temporal adaption of isolates. Moreover, fitness increased in media representing past conditions, providing evidence for past selection for successful growth. These findings were unaffected by limiting our analysis to isolates with vigorous growth rates or to soil obtained from different geographical sites. Spatial structuring, either at large (kilometer) or small (meter) scales, did not significantly influence bacterial fitness, indicating a large role of dispersal in soil bacterial biogeography at these scales. Lastly, we correlated environmental factors such as nutrient ion availability, mean temperature and pH with the obtained fitness landscapes to deduce key factors influencing bacterial temporal adaptation in nature.

POSTERS

D23SY02PS0014

TRANSCRIPTOMIC DIVERGENCE IN RELATION TO MATING SUCCESS OF LEKKING GREAT SNIPES (GALLINAGO MEDIA)

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Sexual selection is a key area in evolutionary biology, explaining the evolution of seemingly maladaptive traits such as conspicuous mating behaviour. Great snipe (Gallinago media) males aggregate on leks, visited by females for the sole purpose of mating, creating the prospect for intense sexual selection. However, the molecular basis of sexual selection in this species is still unknown. In this study, we sequenced the blood transcriptomes of 14 male great snipe individuals with different mating success to identify genes that might be related to sexual selection. One full 454 FLX Titanium run with individually tagged samples was used, and the de-novo assembly of the great snipe transcriptome using all the data consisted of 4476 transcripts (isotigs) with average length of 896 bp, together with 153431 high-quality singletons. The great snipe transcripts were homologous with 4900 chicken genes and 4853 zebra finch genes. Differential gene expression analyses were performed by comparing the sequencing read counts between the mated (9) and unmated (5) individuals, which allowed us to identify 4 differentially expressed genes. In addition, we also identified 2874 single nucleotide polymorphisms (SNPs) and compared the SNP genotype variation between the mated and unmated groups, which led to the identification of 18 genetically structured SNP loci. The functions of differentially expressed genes included 'oxygen transport' and 'viral RNA degradation'. Interestingly, the gene coding for alpha-1-globin was identified as a candidate for both differential expression and for nucleotide divergence. To establish functional correlations between the candidate loci and the ecological variation, these candidate genes need further investigation. To this end, we provide a large number of functional genetic markers (SNPs and SSRs) that can be utilised in follow-up studies.

D23SY02PS0094

NATURAL SELECTION, QUANTITATIVE GENETICS AND BIOENERGETICS OF AN INVASIVE SPECIES: THE LAND SNAIL CORNUASPERSUM

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One of the goals of evolutionary physiology is to understand how natural selection acts, and provokes a response, in functional characteristics of organisms. In this work we measured morphological and physiological components of the multivariate breeder's equation in the introduced populations of land snails.. Terrestrial gastropods are sensitive to water scarcity and are severely energy-constrained. Cornu aspersum snails were recently introduced into Chile, and thus represent an ideal model to investigate contemporary adaptive evolution. Given that populations from the North of Chile (La Serena) experience considerably drier and warmer conditions than populations from the South (Valdivia), we predicted relaxation of selection and accumulation of additive genetic variance in the South. We performed a quantitative genetics experiment in which about 30 half-sib families (second generation), in each population were bred and physiological, life-history and morphological traits were recorded. Additionally, we performed a natural selection experiment where about 600 measured (and lab-born) individuals, derived from the three populations (F3) were released in replicated enclosures at the two extreme locations. Individual fitness (survival) was measured and survival curves constructed. We found very low additive genetic variation/covariation in most traits, including morphological, and high non-additive/maternal effects. The shape of selection was remarkably similar in both extreme populations, irrespective of origin and climate. The best glm model (by AIC) dropped all traits but body mass and standard metabolic rate (residuals), which appeared combined in correlational selection in the North, and with directional/stabilizing selection in the South. These results support the idea of energetic constraints to evolution, irrespective of climate and origin, but the absence of additive genetic variation even in morphological trait is intriguing.

D23SY02PS0101

EVOLUTION OF NATURAL MICROBIAL POPULATIONS IN TRADITIONAL FERMENTED PRODUCTS

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I will present results on the evolution of microbial communities in traditional fermented products from Zambia. These products are produced by transferring a fraction of a former batch to initiate a new batch of product. In this way, microbial communities are allowed to evolve of large numbers of generations. Local villagers maintain independent replicate product lineages. Methods. I have sampled three different traditional products (based on either milk or maize) from across Zambia, obtaining a total of 36 samples. I have established the community composition of the products and compared this composition for different samples of the same product type and different product types. In the laboratory, we have propagated the microbial communities for an additional two months under various conditions and have monitored changes in community composition. Results. Our results show that the microbial flora is dominated by around 6 species of lactic acid bacteria and that specific combinations of species result into stable communities. I show that both geography and anthropogenic factors affect community composition in products collected from Zambia. Laboratory experiments propagating communities over two months show specific shifts in community structure that is consistent over all nine independent communities. Conclusions. Traditional fermented products are very powerful experimental systems to study long-term properties of entire microbial communities. From the field sampling study we conclude that both environmental factors (that differ by geography) and anthropogenic factors are key in shaping microbial communities. From the laboratory studies we conclude that these communities are very stable in the long-term. This work opens up a range of possible follow-up projects. We will perform manipulative experiments in the field as well as in the laboratory to establish exactly what factors are essential for community stability.

D23SY02PS0134

framework.

USING BOTH DIVERGENCE AND ALLELE FREQUENCY INFORMATION IN A UNIFIED FRAMEWORK FOR SWEEP DETECTION

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SweepFinder is a commonly used tool to scan for selective sweeps that are fixed or nearly fixed. Currently, it uses only information contained in polymorphisms in the vicinity of the selected locus. Several recent studies have concluded that including information from invariant sites increases power to detect and localize sweeps. However, this approach also leads to false positive signals in regions that have low mutation rates compared to the background, or that are subject to strong purifying selection. Here, we explore the possibility of instead including information from a third class of sites, fixed differences from an outgroup species, into the composite likelihood framework. This approach is similar to that of the Hudson-Kreitman-Aguadé (HKA) test, but also incorporates the spatial pattern of polymorphism around the putatively selected site. By testing the method with simulations, we find that using only the information from fixed differences and polymorphic sites yields the same power as incorporating information from all sites, without increasing the false positive rate in regions with low mutation rates. We also demonstrate that including fixed differences is more robust to strong and recent bottlenecks than the method that uses only polymorphic sites. Finally, though robustness to bottlenecks is in general increased by our approach, recent bottlenecks with intermediate strength still yield a high frequency of false positives. Here we provide a powerful tool to scan for selective sweeps that incorporates the spatial patterns of the site frequency spectrum and diversity reduction in one single

D23SY02PS0140

SEXUALLY ANTAGONISTIC GENES IN A NATURAL POPULATION OF AFRICAN BUFFALO

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The view is emerging that intralocus sexual conflict at so-called sexually antagonistic genes is a fundamental factor for the genetic architecture of fitness. However, we do not know of any study on a natural population which has been able to analyse sexually antagonistic genes directly. Here, we analysed heterozygosity-fitness correlations (HFC) using microsatellites to show the occurrence of sexually antagonistic genes in the African buffalo (Syncerus caffer) of Kruger National Park. Additional analyses provided new insights into the characteristics of these genes and the selection pressures involved. Expected heterozygosity increased in low body condition (LBC) females relative to high body condition (HBC) females, i.e. a negative HFC, while the opposite was observed in males, i.e. a positive HFC. Alleles with a high frequency in HBC relative to LBC females tended to have a low frequency HBC relative to LBC males. This observation indicates that the particular group of sexually antagonistic alleles linked to the studied microsatellites are beneficial to females and deleterious to males. Furthermore, sexual antagonism was strongest among the high frequency alleles, indicating that they are under positive selection. The sexually antagonistic alleles were dominant in females, indicated by LBC-HBC allele frequency differences among heterozygotes, but less dominant and possibly recessive in males. This sex-specific inheritance pattern may result in protected polymorphism. Pregnant females with relatively many sexually antagonistic alleles, expected to be beneficial to their daughters, were mostly carrying female foetuses, while those with relatively few sexually antagonistic alleles mostly male foetuses. Thus mothers try to improve the chances that sexually antagonistic alleles are transmitted to the sex they benefit. This shows that the genetic load of sexually antagonistic variation can be high enough to interfere with the sexual selection of good genes.

D23SY02PS0189

WHOLE-GENOME METHYLATION PATTERNS IN THE KHOMANI BUSHMEN OF SOUTH AFRICA

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The Khoe San hunter-gatherers of southern Africa are believed to be one of the world's most ancient populations based on their high genetic diversity and low levels of LD. Phenotypically, the Khomani Bushmen have a range of lighter skin pigmentations, intermediate between European and central African populations, as well as a typically shorter stature. We have collected genetic and phenotypic information from the Khomani Bushmen. Moreover, we methyl-typed these individuals at over 400,000 CpGs sites as well as genotyped them at over 300,000 SNP sites. In order to better understand the epigenetic contribution of DNA methylation to skin color, we use this data set to study genomewide methylation profiles for skin pigmentation. We also study the relationship of DNA methylation and age in these individuals. These results can help us better understand the role of DNA methylation in the aging process and in shaping the human phenotype.

D23SY02PS0202

A POTENTIAL ROLE FOR ECOLOGICALLY MEDIATED SEXUAL SELECTION IN THE DIVERGENCE OF TROPICAL PACIFIC HONEYEATERS (MYZOMELA)

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The relative roles of neutral and selective processes in divergence are of key interest in evolutionary biology. Due to the many large and small islands, the Indopacific area constitutes an ideal natural laboratory to study the different modes of diversification. It is a biotically extremely rich region, and as a result is designated as several distinct biodiversity hotspots. It was suggested that the islands of the Pacific are a likely source of biodiversity, rather than a sink of species that originated on the mainland, as has long been the established hypothesis. We use landscape genetic approaches to study the potential roles of drift, natural selection, and sexual selection to study the divergence among island and mainland species of sexually dimorphic honeyeaters, Myzomela sp. Sequence and microsatellite data indicate that these populations either very recently diverged or experience ongoing gene flow, and planned coalescence-based analyses will be useful in distinguishing these two hypotheses. We found that males exhibit distinct differences between islands in plumage coloration and song, but not in other, fitnessrelated morphological traits. Females did not show divergence in any morphological traits between islands. Environmental heterogeneity explained the observed divergence in song frequency characteristics, whereas oceanic barriers better explained divergence in temporal traits. These results suggest that sexual selection is a potential factor in maintaining and deepening population differentiation, and that environmental conditions pose selection pressures on some but not all sexual traits. The role of sexual selection in population divergence will be further evaluated using mate choice experiments.

D23SY02PS0215

SEXUAL SELECTION AND AUTOSOMAL VERSUS SEX-LINKED INHERITANCE OF COLOUR ORNAMENTATION IN THE BLUE TIT

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In its most traditional form, sexual selection has been classically considered as an asymmetrical process whereby choosing partnership is incumbent upon females and elaborating attractive sexually selected traits incumbent upon males. However, the theory of sexual selection is presently going through a major paradigm shift towards considering how sexual selection affects the evolution of female traits. In species with low sexual dimorphism, extravagant female traits are hypothesized to result either from indirect selection acting on male traits, and/or direct selection on female traits. The present study used data on Mediterranean populations of blue tits *Cyanistes caeruleus* over 8 years to explore selection on and genetic (co)variations in male and female coloration. Blue tits are slightly sexually dichromatic birds with females presenting a decreased coloration in the UV-blue cap but not in the yellow chest. Several studies in males, and more recently in females, suggest that these two traits have an important signaling function in the context of sexual selection. Based on 3000 samples of feathers we analysed 8 spectral measures describing the chromatic and achromatic blue cap and yellow chest characteristics. Long term pedigrees allowed us to estimate autosomal inheritance for these traits in males and females, but also the largely ignored sex-(Z-)linked inheritance. For all traits that displayed additive genetic correlations in both males and females, cross-sex genetic correlations tended to be strong, as predicted by the correlated response hypothesis. Despite some correlations between fitness components and colour, selection gradients were weak overall and varied between populations. This study adds to the slowly growing number of investigations on the role of sex-chromosome linkage and cross-sex genetic correlations in the evolution of sexual ornaments and sexual dimorphism. It also reveals betweenpopulation differences in selection and inheritance.

D23SY02PS0264

MATING PATTERNS AND SEXUAL SELECTION IN A SNAPDRAGON HYBRID ZONE

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Large scale, long term evolutionary processes such as gene flow, drift or species formation depend ultimately on the patterns of mating between individual organisms. Mating patterns are especially interesting in natural hybrid zones, where alleles from parental populations recombine to form novel genotype combinations. In the Spanish Pyrenees, one yellow- and one magenta-flowered subspecies of the snapdragon Antirrhinum majus form such a hybrid zone, leading to floral colours not seen in either subspecies. Complementing a broader study to assign parentage across generations, I am genotyping wild seedlings to assign paternity to candidate fathers. This information can be used to infer the ways in which pollinators mediate the dispersal of gametes, and the effects of multiple mating on the distribution of siring success of different fathers with a single mother.

D23SY02PS0350

CROSS-CONTINENTAL VARIATION IN SEXUAL SELECTION AND ITS EFFECT ON THE CONTRASTING REVERSAL OF SEXUAL SIZE DIMORPHISM IN CLOSELY RELATED SEPSID FLY SPECIES (SEPSIDAE: DIPTERA)

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Sexual size dimorphism (SSD), arises when the net effects of natural and sexual selection equilibrate differently in males and females. Species and even populations within a species can exhibit quantitative variation in the extent of SSD, but qualitative reversals in SSD are rare. In a previous study, we documented that sexual selection accounts for a geographic reversal of SSD in the widespread sepsid fly Sepsis punctum. Here, we study the variation in sexual selection acting on male body size in three other closely related species that display contrasting reversals of SSD in Europe (EU) and North America (NA). In all three species, laboratory common garden experiments indicate clear genetic differentiation among populations in body size and development time. Populations of S. cynipsea display female-biased SSD. In its sister species S. neocynipsea, females are larger in EU, whilst NA populations exhibit male-biased SSD. This is a sharp contrast to *S. punctum*, in which NA populations display female-biased SSD, whereas in EU males are the larger sex. Yet in another species, S. biflexuosa, SSD was consistently absent, with both sexes having similar body sizes in EU and NA. Next, we conducted mating experiments under three increasing male-biased operational sex ratios. Although sexual selection intensity on male body size based on pairing success was overall positive, contrary to *S. punctum* there was no correlation between the intensity of sexual selection and male body size in these species. It is possible that the effects of viability selection acting on the sexes and/or fecundity selection on the female could be stronger than sexual selection on males, which requires further investigation.

D23SY02PS0366

PHENOTYPIC DIVERGENCE AMONG WEST EUROPEAN POPULATIONS OF REED BUNTING EMBERIZA SCHOENICLUS AGREES WITH MIGRATORY AND FORAGING BEHAVIOURS INDICATING LOCAL ADAPTATION

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Divergent selection is responsible for many phenotypic differences between populations, potentially leading to speciation through the evolution of reproductive barriers. Here, we evaluated the morphometric divergence among west European populations of Reed Bunting (Emberiza schoeniclus) in order to determine the extent of local adaptation relative to two important selection pressures often associated with speciation in birds: migration and diet. We also compared the winter diet between the subspecies using stable isotope analyses. As expected by theory, migratory schoeniclus had longer and more pointed wings and a slightly lower body mass than the resident subspecies, with the exception of lusitanica, which despite having rounder wings was the smallest of all subspecies. Tail length did not vary according to the expectation (shorter tails in migrants) probably because it is strongly correlated with wing length. The southern, resident subspecies showed a narrower foraging niche clearly different from the migratory subspecies. The isotope signatures are consistent with previous anecdotal observations that resident birds feed on insects lying dormant inside the reed stems, whereas migratory birds feed on seeds of which C4 plants are strongly represented. Accordingly, the resident witherbyi had a very thick, stubby bill and, despite being smaller, the resident lusitanica had a significantly thicker, longer bill than migratory schoeniclus, which had a thin bill. Furthermore, geometric morphometric analyses revealed that the southern, resident subspecies have a more convex culmen than the migratory schoeniclus, being able to exert a greater strength at the bill tip, which is in agreement with their feeding technique. Thus, the three subspecies occurring in Western Europe differ in a variety of traits following the patterns predicted from their migratory and foraging behaviours, strongly suggesting that these birds have became locally adapted through natural selection.

D23SY02PS0367

POPULATION GENOMICS OF A SPREADING EXOTIC PLANT SPECIES

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Human activities increasingly introduce plant species far outside their native range, in regions where they did not evolve, under environmental conditions to which they might be poorly adapted. Therefore, before becoming invasive, exotic plant species may rapidly evolve in their new environment in response to the prevalent adaptive forces. To evaluate the possibility of genetic adaptation preceding invasive spread, we performed a population genomic study of Sisymbrium austriacum subsp. chrysanthum populations (the Pyrenean Jeweled rocket, Brassicaceae). The species is native to the Pyrenees but has been documented in Belgium, 1200 km North of its native range, since the 19th century. To discover SNPs, a subset of individuals was screened using Paired End RAD-Seq. The native and both current and historical non-native gene pool were then screened on a set of SNPs in genes associated with ecologically important plant traits. FST outlier analysis combined with gene function enrichment analysis identified candidate SNPs under positive selection in major floweringtime (FT) Arabidopsis homologs. Time series supported selection from standing variation in FT genes during the initial phases of colonization.

D23SY02PS0377

UNEXPECTED BENEFIT OF THE SOCIAL PARASITE MICRODON MUTABILIS FOR A KEY FITNESS COMPONENT OF ITS ANT HOST

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Numerous invertebrates inhabit social insect colonies, including the hoverfly genus *Microdon*, whose larvae typically live as brood predators. *Microdon mutabilis* show a remarkable pattern of local adaptation to their host *Formica lemani* and ant colonies apparently endure *M. mutabilis* infections over several years despite losing a considerable fraction of their brood to *Microdon*. In fact, empirical evidence suggests that infected colonies may even produce more gynes. From an ecological and evolutionary perspective this constitutes a puzzle. Here we present a model for resource allocation within polygynous ant colonies, which assumes that whether an ant larva switches development into a worker or a gyne depends on the quantity of food received randomly from workers. Accordingly, *Microdon* predation promotes gyne development by increasing resource availability for surviving brood. Several model predictions are supported by empirical data. (i) Uninfected colonies rarely produce gynes at all. (ii) Infected colonies experience a short-lived peak in gyne production leading to a bimodal distribution in gyne production. (iv) Parasite induced reduction in the brood:worker ratio is the critical mechanism controlling gyne production to become noticeable.

D23SY02PS0380

CAN A HYBRID OUTCOMPETE ITS PARENTAL SPECIES? - A STORY ABOUT A "SUPER CLONE" (DAPHNIA GALEATA × LONGISPINA HYBRID)

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The role of hybridization in evolution has been underestimated for a long time. For example, hybrids were often considered less fit then their parental species, due to genetic incompatibility. However, hybridization occurs frequently in plants and in animals. Our recently collected field data showed that a Daphnia community of a small quarry lake in Munich, normally consisting of parental species and hybrids belonging to the *D*. *longispina* complex, has become dominated by a single hybrid clone - the "super clone". By creating artificial communities consisting of the "super clone" and other clones of the D. longispina complex, we proved the competitive strength of the "super clone". After ~6 generations the "super clone" had increased from 8% to 100% in some of the artificial communities. Additionally, we studied fitness parameters of these clones, kept under two different temperatures. Here, there was no special performance of the "super clone". Therefore we are currently comparing the carrying capacity of "super clone" with the other clones, by measuring the change in numbers of Daphnia over time, in a certain volume of water. Additionally, we will study fitness parameters of the clones under crowding (and control) conditions. The crowded media will be obtained from crowded stock cultures. We expect the "super clone" to have higher carrying capacity as well as to be less affected by crowded media than the other clones, thereby preventing the other clones from reproducing and establishing within the population. Finding the pattern that makes the "super clone" so special will contribute to a general knowledge about hybrid-specific traits and their contribution to evolutionary processes.

D23SY02PS0390

WHY DO INBRED ANTS LIVE LONGER?

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Inbreeding has profound consequences on a genomic, individual, population and even species level. However, the effects of inbreeding on natural populations, especially in insects, remain largely unstudied. Ants are an ideal model system to study the consequences of inbreeding, as they are ecologically important and form socially complex groups. Social groups provide another level possibly affected by inbreeding, as the large amount of interactions between numerous individuals could give rise to socially mediated inbreeding depression. Inbreeding has been shown to have negative consequences both at the individual and colony level in a natural population of the narrow-headed ant (*Formica exsecta*). Here, we compare the resistance of inbred and outbred worker ants against starvation. Our results indicate that inbreeding has no effect on longevity under starvation, however, naïve (fed) inbred ants outlive the outbred ones. What makes inbred ants live longer? We investigated these questions using both behavioural assays and gene expression studies.

D23SY02PS0404

SIGNAL DESIGN AND PERCEPTION IN IBERIAN REED BUNTINGS

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Song divergence in different populations of a species can lead to reproductive isolation and speciation. Some passerines have different singing styles for distinct social contexts and songs from each style change over time and space at different rates. Song divergence between subspecies should therefore differ with singing style. The reed bunting (Emberiza schoeniclus) has three singing styles: Fast, related to obtaining a social mate; Dawn, related to extra-pair paternity; and Slow, used as an all-clear signal to social females. We compared properties of songs from the three styles among populations of the two resident subspecies breeding in southwestern Europe (lusitanica and witherbyi). In addition, we compared the reactions of territorial males of both subspecies to playback of Fast song from their own and two foreign subspecies (the other southwestern subspecies and the central and northern European schoeniclus). Overall, subspecies differ more in Fast and Dawn songs than in Slow songs, whereas divergence level in Fast and Dawn songs is very similar. Larger subspecies song divergence in Fast and Dawn singing styles is probably due to stronger female preference for local song in these styles (intersexual selection), rather than to greater song sharing and matching between males (intra-sexual selection). Regarding signal perception, both witherbyi and lusitanica males strongly discriminate schoeniclus song. However, witherbyi shows a more consistent discrimination of lusitanica song than lusitanica does of witherbyi song. Differential territorial defense suggests that an intruding male with different songs does not represent a competitive threat and is expected if females mate assortatively with respect to song. Thus, our results provide evidence for a faster evolution of sexually selected songs, and for premating reproductive isolation among reed bunting subspecies.

D23SY02PS0427

POPULATION GENOMIC SCAN FOR NATURAL VARIATION AND SELECTION ACROSS CENTRAL METABOLISM

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A growing number of studies are finding empirical evidence for varying selection dynamics across the enzymes of central metabolism, with steps early in pathways being the focus of positive selection in taxa as divergent as flies and plants. Here we explore these dynamics in the butterfly *Melitaea cinxia*, where two enzymes of central metabolism have been shown to harbour genetic variation correlated with various aspects of flight performance and fitness in the wild. Using a Pool-Seq approach, the site frequency spectrum of polymorphisms in each of two long diverged populations was assessed across central metabolic genes. This variation was then compared between these populations to discover SNPs that displayed significant changes in frequency. Furthermore, through comparison with a sister species, this variation was assessed for selection dynamics, finding evidence for selection upon enzymes early in the studied pathways. Finally, to assess the significance of this variation in extant populations, we compared SNP frequencies between young and old butterflies of a single population, finding SNPs associated with survivorship in the wild. Together, this comparative genomic analysis of SNP variation provides important insights into selection dynamics acting on central metabolism.

D23SY02PS0434

INTER- AND INTRASPECIFIC RELATIONSHIPS BETWEEN PERFORMANCE AND TEMPERATURE IN A CRYPTIC-SPECIES COMPLEX OF THE ROTIFER BRACHIONUS PLICATILIS

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The strategy of decreasing size with increasing temperature operates at geographic (Bergmann's rule) and within-genotype (temperature-size rule) scales and presents a puzzle to researches. The aim of this study was to verify whether differences in the adult size of closely related species coexisting in the same habitat are adaptive. The experiments were conducted on the cryptic species complex of the rotifer Brachionus plicatilis, represented by three species living in the Torrablanca marshes of eastern Spain. A cryptic species complex is a group of species each one satisfying the biological definition of species but having very similar morphologies. It provides an experimental model for testing ecological hypotheses because the species similarity tends to simplify comparisons. The monogonont rotifer species complex Brachionus plicatilis is a good example. Three of the cryptic species co-occur in the same ponds and differ in adult size. Co-occurrence is associated with their different preferences for temperature and salinity, mode of competition and vulnerability to predators. Three aspects of the performance-temperature relationship along a temperature gradient were studied, (i) the population growth rates were compared, (ii) the interspecific phenotypic plasticity of adult size was assessed, and (iii) the pattern of sediment egg bank hatching was explored. The results show that (i) the optimal temperature for population growth is related to species size in a manner foreseen by Bergmanns' rule, (ii) the temperature-size rule differs between species and may depend on the level of temperature specialization, and that (iii) adult size, as a proxy for resting egg size, is adaptive regarding the hatching pattern. These results broaden the general knowledge on the evolutionary mechanisms behind the body size-temperature relationship and have important consequences for the coexistence and geographic ranges of species, and for species redundancy.

D23SY02PS0441

IDENTIFICATION OF X-LINKED LOCI AFFECTING COLD STRESS RESISTANCE IN DROSOPHILA MELANOGASTER BY MEANS OF COMBINED QUANTITATIVE TRAIT LOCI AND SELECTIVE SWEEP MAPPING APPROACHES

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With the aim of understanding the molecular basis of cold adaptation in *Drosophila melanogaster*, we initiated the identification of the genes/functional elements affecting the fly's chill coma recovery time (CCRT). CCRT is a proxy for cold stress tolerance. Based on quantitative trait loci (QTL) mapping between inbred lines from temperate and tropical origin, three X-linked QTL influencing CCRT were identified at 17, 18 and 24 cM. We narrowed them further by means of quantitative deficiency mapping. Plus we gained further resolution, to the gene/nucleotide level, for the locus at 18cM by subjecting the targeted chromosome fragment to population genetic analysis, quantitative mutation mapping and gene expression analyses. In the light of selective sweep theory we expect the alleles responsible for cold tolerance to have been positively selected in the temperate population, therefore regions in the QTL that show footprints of positive selection may contain the candidate molecular elements behind cold stress tolerance. However, selective sweep mapping overlooks sites with other modes of evolution. In oder to cover a broader range of potential sites influencing cold stress tolerance we conducted a parallel search with a phenotype-genotype approach suitable for fine scale mapping of QTL. Within the QTL defined at 18 cM, a 3-kb long transcription enhancer element approximately 7kb upstream of gene *brinker*, shows footprints of positive selection and is strongly associated CCRT. We are currently studying the role of this enhancer element in the phenotype of interest, by measuring expression-level differences of the gene brinker and its immediate neighbor genes, both in wild type tropical/temperate flies and transgenic tropical flies with the putatively selected temperate version of the enhancer.

D23SY02PS0448

COMPLEX GENETIC ARCHITECTURES IN NATURAL POPULATIONS: A SCREEN FOR COADAPTED MITOCHONDRIAL-NUCLEAR ALLELIC INTERACTIONS FOR MALE FERTILITY ACROSS DISJUNCT POPULATIONS

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When it comes to understanding fundamental questions such as how the genotype maps to the phenotype, and how selection influences adaptive evolutionary processes, research has focused on the allelic variation within the nuclear genome. By contrast, effects by variation within the mitochondrial genome have been neglected because they were suggested to be absent or neutral. Recent studies in a range of organisms substantially altered that view and suggest that not only the nuclear, but also the mitochondrial genotype and the interaction between the two affect the phenotype and hence respond to selection. To test our hypothesis, that selection in natural populations affects mitochondrial and nuclear genomes as an entity (i.e., they co-evolve), we established fly lines (*Drosophila melanogaster*) originating from two disjunct natural populations that have evolved under distinct climatic regimes. We introgressed the mitochondrial genome from both populations in the native and the foreign nuclear genotypic background to analyse the effects of mitochondrial and nuclear genotype independently. Theory and recent empirical data show that mitochondrial-nuclear interaction effects should mainly affect males, particularly male reproductive traits. That implies, given that differences in reproductive success are the raw material for natural selection, that mitochondrial genotypes play an important part in evolution. We are now (February to April 2013) running a large scale sperm-competition experiment to examine the relative mitochondrial and nuclear genetic contributions to male fertility, and in particular to screen for complex gene-by-gene-by-environment interactions. The presence of such genetic interactions would likely fundamentally alter the expected evolutionary responses to selection on these male reproductive traits in the wild, particularly when we consider that mitochondrial genomes are maternally inherited and thus unlikely to respond directly to selection on males.

D23SY02PS0449

THE GENETIC ARCHITECTURE OF THE LATITUDINAL VARIATION IN SEXUAL DIMORPHISM OF MEDAKA

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Sexual dimorphism, morphological differences between the sexes, is widespread a throughout the animal kingdom. However, the degree of sexual dimorphism varies considerably among closely-related species or even populations. Although there are many theoretical genetic models that can explain the genetic mechanisms of variation in sexual dimorphism, little empirical studies have been conducted to investigate the genetic basis of the variation in sexual dimorphism among natural populations. How many genes are involved in the variation in sexual dimorphism? Are they localized at particular chromosomal regions, such as sex chromosomes? Are different dimorphic traits regulated by different genes? In the Japanese medaka (Oryzias latipes), populations at lower latitude are more sexually dimorphic than populations at higher latitude. For example, anal and dorsal fins of mature males in low-latitude populations are longer than that of the higher-latitude males. Laboratory rearing experiments revealed that these differences in male fin length is mediated by the difference in the rate of fin elongation during development, in particular after a certain body size which probably represents sexual maturation. Furthermore, males from lower-latitude populations show more frequent fighting and courtship behaviors than males from higher-latitude populations. In this study, genetic basis for these morphological and behavioral differences were investigated using QTL mapping. We crossed two populations of medaka from habitats of different latitude (Aomori and Okinawa) and quantified several traits related to fin elongation process and mating behavior in F2 progenies. We next designed custom SNP assay system to create a linkage map and conduct QTL mapping. We found significant QTL controlling different sexually dimorphic traits on a single autosome, suggesting that these secondary sexual characters may be controlled by the same genetic mechanism.

D23SY02PS0460

DIFFERENTIATION OF PHOTOSYNTHETIC CHARACTERISTICS BETWEEN COASTAL AND FRESHWATER LAKE POPULATIONS OF THE COASTAL PLANT LATHYRUS JAPONICUS (FABACEAE)

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Lake Biwa, formed around 4 million years ago, harbors many coastal species that commonly inhabit seashores. These coastal plants have been reported to exhibit morphological, physiological, and genetic differentiations between Lake Biwa and coastal populations (e.g., Ohtsuki et al. 2011). We also reported the beach pea, Lathyrus japonicus, is a typical coastal species of this freshwater lake, has become those differentiations, suggesting habitat-adaptation to each environment. However, photosynthetic characteristics has been remain unknown nevertheless its importance as survival factor to plants. The present study aimed to measure and compare photosynthetic characteristics between Lake Biwa and coastal transplanted populations using LI6400. In Lake Biwa lakeside, transplanted plants (coastal and lakeshore individuals) exhibited similar pattern of light-photosynthesis-curve. However, in coastal environment, inland individuals showed lower photosynthesis rate than seaside ones. On the other hand of photosynthesis rate, coastal individuals in Lake Biwa lakeside were higher than those in original habitat. In addition, transplanted lakeshore individuals showed low stomatal conductance compared with coastal individuals. Meanwhile, photosynthetic efficiency (Fv/Fm), electron transport rate (ETR), photochemical and non- photochemical quenching rate (qP amd qN) were similar between those populations. Thus, transplanted lakeshore individuals decreased photosynthetic rate, suggesting lack of acclimation to coastal environment. In contrast, transplanted coastal individuals were easily acclimated to lakeshore environment, suggesting their adaptability to both inland freshwater and coastal habitats. The differentiation of photosynthetic characteristics might trigger incipient speciation between inland and coastal populations.

D23SY02PS0468

NEUTRAL AND SELECTION-DRIVEN DECAY OF SEXUAL TRAITS UNDER ASEXUALITY

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Vestigialization of non-functional traits is an important phenomenon in evolutionary biology, highlighting the importance of natural selection for adaptations. Both drift and selection can drive trait decay, and disentangling the influence of these processes for specific traits has proven to be challenging. The fate of sexual-reproduction traits in asexual all-female lineages can provide insights into both, vestigialization via drift and selective processes, because of the many replicate events independently leading from sexual ancestors to asexual lineages in diverse taxa. In addition, because certain traits, notably male-specific traits, are not exposed to selection under asexuality, these traits can only decay as a consequence of drift, while other traits would be largely under negative selection. We review the fate of male and female sexual traits in 78 animal species characterized by asexual reproduction, covering a broad taxon range including nine different insect orders, arachnids, mites and molluscs. Most asexual lineages are still able to rarely produce males and these asexually-produced males are often largely or even fully functional. By contrast, for asexual females there is convergent and rapid vestigialization of sexual traits, especially of traits related to mate attraction and location which are expected to be associated with costs. The level of decay of certain female traits, in addition to asexual females being unable to fertilize their eggs, would severely impede reversals to sexual reproduction, even in recently derived asexual lineages. Overall, the convergent maintenance vs decay of different trait types across diverse asexual lineages suggests that neutral traits display little or no vestigialization even after extended periods under relaxed selection, while extensive vestigialization for (presumably) selected traits occurs extremely rapidly.

D23SY02PS0471

LINKING PHENOTYPE TO TRANSCRIPTOME: SEXUAL DIMORPHISM AND SEX-SPECIFIC SELECTION IN BIRDS WITH DIFFERENT MATING SYSTEMS

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We assembled de novo transcriptomes for six species within the Galloanserae (fowl and waterfowl) in order to assess the link between complex phenotypic sexual dimorphism and global gene expression differences between males and females. The six species exhibit the full range of sexual dimorphism, from monomorphism observed in geese to extreme sexual dimorphism in peafowl. This gradient of sexual dimorphism within a single clade makes it possible to directly test how sexual selection affects genome evolution and how sexually dimorphic phenotypes are encoded. To do this, we generated an extensive RNA-Seq dataset, with on average 95 Gb of next-generation Illumina sequencing data for each species, sampling both male and female spleen and gonad tissue. After constructing de novo transcriptome assemblies for each species using Trinity and establishing orthology across all six study species, we assessed sex-specific gene expression and sequence evolution of sex-biased genes in order build a comprehensive picture of the role of mating system in genome evolution. The generated de novo transcriptomes for each species contain on average 15,847 significantly expressed contig groups, which resulted in 12,337 predicted peptides on average per species, from which orthology was inferred across the clade. Our results indicate that transcriptional masculinization and feminization are correlated with the overall degree of sexual dimorphism in phenotype, and that sex-specific selection shapes coding sequence evolution of sex-biased genes.

D23SY02PS0495

VARYING PATTERNS OF GENETIC CORRELATIONS BETWEEN MORPHOLOGICAL TRAITS ALONG THE ONTOGENY OF BLUE TIT NESTLINGS

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Morphological traits usually are strongly genetically determined, which results in relatively high heritabilities of such traits compared to life-history traits. However, apart from their genetic background, different phenotypic traits may be correlated at the genetic level with other traits. Such correlations - if strong - may hamper or strengthen evolution of pairs of traits, depending on the direction of genetic correlations. Here we analyse morphological traits such as body weight and structural body size from a long-term study of the wild population of blue tits. We measured morphological traits during the early development of nestlings at several points in time. By employing animal models we were able to estimate genetic and non-genetic sources of variance in these traits and covariances between different traits. We demonstrate that the magnitude of correlations between analogous traits expressed at different ages. Our results indicate that traits expressed during development are not genetically uniform but have their internal, complex quantitative genetic architecture.

D23SY02PS0521

THE HOTTER THE BETTER: TEMPERATURE INDUCED DIFFERENCES IN GROWTH AND MORTALITY

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With increasing global temperatures knowledge about how organisms may cope with different thermal environments has become of paramount interest. Lake Mývatn on northern Iceland provides a unique setting for studying temperature adaptation. Within the lake there are several cold (6°C) and warm (23°C) geothermal springs over a relatively short distance, creating temporally stable thermal regimes separated by areas of seasonally varying temperatures. I investigated temperature adaptation in populations of a pulmonate snail (Radix balthica) sampled throughout the lake. A reciprocal transplant experiment conducted in the field and a common garden experiment performed in the laboratory (F3 generation) showed that snails from warm localities always grow faster than cold-origin snails. In addition, mortality was higher for cold originating snails in a warm environment compared to warm originating snails in a cold environment. Thermal preference and tolerance (tested on F1 generation) were not different between the four extreme temperature populations or when grouped according to thermal environment. Molecular analyses (AFLP) showed substantial differences corresponding to the two thermal extremes within the lake. My findings support co-gradient temperature adaptation model and suggest that temperature variation can act as a strong selective barrier within an open aquatic system.

D23SY02PS0530

DIFFERENCES IN MTDNA COPY NUMBER BETWEEN INDIVIDUALS WITH LONG AND SHORT HAPLOTYPES FROM A NATURAL POPULATION OF DROSOPHILA SUBOBSCURA

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In the absence of meiosis mitochondrial DNA (mtDNA) could act as a selfish genetic element, i.e. increase its own fitness at the expense of the fitness of its host. Theoretical models suggest that uniparental transmission of mtDNA in almost all eukaryotes has been evolved to prevent the spread of selfish mutations in the population. A selfish mutation in mtDNA could be a large deletion that reduces the number of genes in the mutated molecule. Hence, this mtDNA, should be less functional but should replicate faster than the non-mutated mtDNA. This example, which is supported by some experimental data, assumes that mtDNA replication is inversely correlated with the length of the molecule and directly correlated with time. According to this model smaller molecules replicate faster than larger ones and produce more copies per unit of time. We have investigated this assumption using a natural population of Drosophila subobscura from Serbia where we found an mtDNA haplotype which is longer by 2.7kb (17%) than the standard haplotypes. Restriction site analysis and partial sequencing showed that long haplotype contains additional A+T rich region, srRNA gene, three specific tRNA genes and a sequence that is combined of ND2 gene and lrRNA gene. Experiments on fitness analysis indicate that this long mtDNA haplotype decreases the fitness of its bearers. We counted mtDNA copy number in same aged flies to investigate whether individuals with longer haplotypes contain less copy number than individuals with standard haplotypes as anticipated by the theoretical model.

D23SY02PS0534

NEWS ON THE NATURAL ECOLOGY AND EVOLUTION OF THE MODEL NEMATODE C. ELEGANS

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Although the nematode *Caenorhabditis elegans* is a major model organism in diverse biological areas and well studied under laboratory conditions, little is known about its ecology and evolutionary history. Therefore, characterization of the species' natural habitats should provide a new perspective on the otherwise well-characterized biology and life-history of this nematode. *C. elegans* was for a long time thought to be a soil nematode, but actually seems to prefer nutrient- and microorganism-rich substrates. In order to extend these findings, this project focuses on a continuous long term sampling of *C. elegans* in rotting apples and compost heaps. Since these habitats degrade rapidly and are only available temporarily, nematodes need to escape harsh conditions and food limitation. We observed that slugs and isopods are likely vectors for transport to new environments. Moreover, *C. elegans* was found to share its habitat with the related nematode species *Caenorhabditis remanei*, which could thus represent an important competitor for a similar ecological niche. Microsatellite markers are currently used to characterize population genetic differentiation of the recently isolated *C. elegans* and those isolated in 2002 from the same location.

D23SY02PS0542

GENETICS OF PARALLEL PHENOTYPIC EVOLUTION IN NINESPINE STICKLEBACKS (PUNGITIUS PUNGITIUS) – A COMPARATIVE GENOMIC APPROACH

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Uncovering the genetic architecture of local adaptations is a central topic in contemporary evolutionary biology. Parallel phenotypic and physiological differentiation in independent populations provides strong evidence for evolution by natural selection. Pond populations of Fennoscandian ninespine sticklebacks, subject to strong genetic drift show parallel phenotypic divergence from their marine conspecifics in several traits (e.g. growth, behavior, body shape) apparently as a result of adaptation to reduced predation-risk. We are investigating genetic mechanisms underlying this parallel phenotypic evolution with the aid of genomic approaches utilizing F2-intercrosses between pond populations and a marine population. One of the main aims is to clarify whether the same (parallel evolution) or different (convergent evolution) genes and genomic regions are responsible for the repeated evolution of pond phenotypes. Targeting this, a high density linkage map based on SNP-markers has been constructed to identify genomic regions responsible for the phenotypic divergence among pond and marine populations. This also opens opportunities for comparative genomic studies among nine- and threespine sticklebacks. In future, linkage and QTL mapping for multiple crosses will give the first insights into genomic regions responsible for parallel phenotypic evolution in ninespine sticklebacks.

D23SY02PS0573

EVIDENCE OF CYTO-NUCLEAR EPISTASIS IN DROSOPHILA SUBOBSCURA

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Cyto-nuclear fitness interactions within population are important in maintaining polymorphism in mtDNA. Proteins of the respiratory chains of eukaryotes are coded from both mitochondria and nucleus. Additionally, genes for mtDNA replication and transcription are coded from nucleus. Despite the tight cooperation, the two genomes evolve at different rates, with mtDNA showing higher mutation rate. The compensatory model predicts positive selection on mutations in nDNA that compensate deleterious mutations in mtDNA, where the natural selection is week mechanism due to maternal inheritance, lack of recombination and small effective population size. Drosophila subobscura shows geographical homogeneity of mtDNA variability, with two dominant haplotypes (I and II) and a series of haplotypes that appear at low frequencies. Previous research indicates that mtDNA in *D. subobscura* has a significant effect upon hosts' fitness, and that fitness differences between the two most frequent haplotypes are due to cytonuclear interactions. One rare haplotype (I-L) that bears large duplication in mtDNA, and has same restriction pattern as haplotype I was found in a natural population. In our present research mtDNA effect on fitness is analyzed by comparing egg-to-adult viability and longevity of mtDNA haplotype I-L with that of the two most frequent haplotypes (I and II). This is done on the nuclear backgrounds of both the I-L haplotype and the two most frequent haplotypes. The results indicate that a large duplication in mtDNA significantly compromises fitness. A significant cytonuclear interaction is observed only when fitness of haplotype I-L is compared to that of haplotype II, but not when fitness of haplotype I-L is compared to that of haplotype I. The results suggest an absence of nuclear compensatory mutations for this duplication in mtDNA, but highlight the importance of cyto-nuclear interactions in maintaining the two most frequent mtDNA haplotypes in *D. subobscura*.

D23SY02PS0574

UNLOCKING POOR QUALITY DAPHNIA SAMPLES BY SNP GENOTYPING

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In order to track temporal-evolutionary changes in the plakton communities, as an important issue remains the proper identification of species and their hybrids. For the analyses of Daphnia longispina complex, microsatellie genotyping is a commonly used method. However, we found these length-based markers unsuitable when analyzing poor quality samples. Therefore the historical, formaldehyde preserved samples remain inaccessible. To overcome this problem, we propose SNP based genotyping, due to possibility of shorter fragment amplification. Furthermore, this method allows not only highthroughput genotyping, but the calibration among laboratories is also relatively precise. Therefore, we aim to develop a reliable method to identify species of the D. longispina complex and their hybrids by SNP genotyping. By comparing the transcriptome of D. galeata with D. pulex genome (wfleabase.org) we are identifying genes and their chromosomal location in order to obtain multi-loci markers, and corresponding primers are then being designed. After the sequencing and alignment of these genes for each species in the complex (D. cucullata, D. galeata and D. longispina), candidate SNPs are being identified. For the small scale confirmation of the diagnostic value of these candidate SNPs, we are sequencing a set of genetically well-defined clones from species and hybrids originating from diverse locations across Europe. For the large-scale screenings we are optimizing multiplex PCR reaction of short amplicons and SNP detection via SnaPshot Multiplex kit. To validate the results, we are applying the developed assay for the samples, which were previously analyzed with microsatellite markers. By multi-locus SNP genotyping we will be able to assess the population structures in long-term formaldehyde preserved samples of a hybridizing species complex.

D23SY02PS0606

ASSORTATIVE MATING STRENGTH AND SELECTION OF MORPHOLOGICAL CHARACTERS IN NATURAL POPULATIONS OF TWO CLOSELY RELATED DAMSELFLY SPECIES

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Assortative mating occurs when individuals in a population mate non-randomly and there is a correlation with respect to traits between individuals in mated pairs. It is important in evolutionary processes in that if selection acts on the same trait, assortative mating may lead to speciation and reproductive isolation. On the other hand, if assortative mating is free of selection, it will lead to stabilizing selection within a population. In this study, I examined assortative mating strength within and between two sympatric damselfly species (Calopteryx splendens and Calopteryx virgo) by correlating the male and female morphological characters in mated pairs. In addition, I investigated the relationship between assortative mating and sexual selection (linear, β and quadratic, γ) on the same traits for these damselfly species. In both species, positive assortative mating was more common than negative assortment (disassortative mating). Thorax width in both species had the greatest assortative mating strength. There was no relationship between assortative mating strength and linear selection or quadratic selection. This indicates that there is no strong connection between assortative mating and sexual selection on the same traits. Assortative mating strength for *C. splendens* and *C. virgo* were concordant, and positive, suggesting that there is no noticeable reproductive isolation between these two species and that stabilizing selection is probably operating. Rather, assortment in both species is based on quality, where high-quality males get high-quality females. This study contributes to the knowledge about evolution and selection in natural populations.

D23SY02PS0615

INBREEDING DEPRESSION IN MALE PRIMARY SEXUAL CHARACTERS: A META-ANALYSIS AND AN EMPIRICAL STUDY IN WILD SONG SPARROWS

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Inbreeding, defined as reproduction among relatives, reduces offspring fitness in many plant and animal species. Such 'inbreeding depression' is a potent force that can shape the evolution of mating systems and drive populations to extinction. It is now clear that inbreeding can depress individual reproductive fitness in a wide range of species in nature. However, despite being critical to predicting consequent evolutionary and population dynamics, the life-history stages and mechanisms that contribute to inbreeding depression in overall reproductive fitness are still little understood. Specifically, the impact of inbreeding on male primary sexual traits, which are key components of reproductive fitness, is not yet clear. We first undertook a qualitative meta-analysis of published data to estimate the direction and magnitude of inbreeding depression in male sperm and pollen performance, and hence in key primary sexual traits in male animals and plants. Second, we estimated inbreeding depression in sperm performance using a wild pedigreed population of male song sparrows in which inbreeding is widespread. Our meta-analysis showed weak inbreeding depression in sperm and pollen performance-related traits. However, there was substantial variance in inbreeding loads across studies and a notable absence of data from wild populations. We then show that in wild song sparrows, sperm velocity and motility, two main predictors of male paternity success, did not vary with individual inbreeding coefficient. This result contrasts with the meta-analysis of non-wild populations, and is unexpected because there is substantial inbreeding depression in male reproductive fitness in the study population. Further studies quantifying the magnitude of inbreeding depression in male primary sexual characters in wild populations, and the corresponding contribution to inbreeding depression in male reproductive fitness, are therefore required.

D23SY02PS0640

MOLECULAR EVOLUTION IN THREE POLYCOMB GENES REVEALS A LOW LEVEL OF VARIATION IN THE NONRECOMBINING DOT-LIKE CHROMOSOME OF D. SUBOBSCURA

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The Polycomb group (PcG) proteins are epigenetic repressors highly conserved from yeast to plants and animals. Clustered in at least three main functional complexes (PRC1, PRC2 and PhoRC) Polycomb proteins are involved in many biological processes, being the silencing of the Hox genes during the Drosophila embryonic development their most studied role. Here, we analyze the divergence of the *Pho* and *Sfmbt* genes coding the subunits of the Pleiohomeotic Repressive Complex (PhoRC) in the 12 Drosophila species with whole sequenced genomes and in three additional species of the obscura group (D. subobscura, D. madeirensis and D. quanche). The divergence study has been extended to *Phol*, a paralog of *Pho*. The multiple alignment of each gene in the 15 species was used in maximum likelihood analyses with PAML to try to detect evidences of selection acting in particular lineages. Finally, nucleotide polymorphism of the three genes has been analyzed in 15 lines of D. subobscura in order to estimate their level and pattern of variation and to detect putative deviations of neutrality. An extremely low level of variation was detected at Pho and the HKA test revealed a significant decoupling between the levels of polymorphism and divergence in this gene relative to the other studied genes. The location of Pho in the dot-like chromosome (Muller's F element) with no, or very low, recombination may account for this result. Indeed, a low variation was previously reported in the F element of the *melanogaster* subgroup species. However, this is the first time that nucleotide polymorphism in genes of the dot-like chromosome is studied in *D. subobscura*, confirming a low level of variation in this nonrecombining chromosome in a species of the *obscura* group.

D23SY02PS0642

COMBINING DEMOGRAPHY WITH QUANTITATIVE AND POPULATION GENETICS TO INFER THE ADAPTIVE POTENTIAL OF SMALL POPULATIONS

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The adaptive potential of small and isolated populations is a central question in evolutionary biology. The ability of small populations to respond to new selective pressures is often limited by low additive genetic variability at loci underlying important life-history traits. These traits are often density- and frequency-dependent, and vary in both space and time. To improve ability to predict the magnitude and rate of adaptation to a changing environment in small populations requires combining demography with quantitative and population genetics. Marble trout (Salmo marmoratus) living in Slovenian streams provide a model system for investigating life-history evolution, adaptive potential and the contribution of stochastic events to population dynamics in small and geographically-constrained fish populations. Marble trout populations are isolated, have low density and small population sizes, and harbor low neutral genetic variation; they also experience high mortality caused by flash floods that further contribute to the erosion of genetic variability. To tease apart the contribution of plasticity, resource limitation, genetic drift and natural selection on expression of life-history traits in marble trout we used theoretical insights, long term mark-recapture data sets, and pedigree reconstruction with panels of SNPs discovered using Next Generation Sequencing-generated data that overcome the obstacles posed by low heterozygosity in marble trout. The reconstruction of multi-generation pedigrees and the long-term mark-recapture data allowed us to: (i) infer mating patterns and average family size; (ii) infer heritability of life-history traits, while controlling for maternal effects and population density; and (iii) test for associations between fitness-related traits and particular genotypes at the individual and family level.

D23SY02PS0660

DISTORTION OF THE SEX RATIO IN FOREST PESTS: AN ECOLOGICAL ALTERNATIVE FOR SELF-REGULATION OF POPULATIONS

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According to the theory of natural selection, there is an equal proportion of sexes. However, this rule is not always an ideal situation, because a higher proportion of one sex can also lead to some benefits such as ensuring fertilization of all females, encourage the dispersal of the species and control the size of population. Epidemics caused by forest pests are endangered trees, thus making less available resources. The scarcity of resources may affect the fitness of females due to higher metabolic and reproductive costs. The objective of this work is to determine if there is a distortion of the sex ratio in conditions of nutritional stress in the spruce budworm, one of the most problematic forest pests in Canada. Three colonies of spruce budworm have been reared for three generations under two types of artificial diets: a control diet and a stress sugar nitrogen. The variables considered are differential mortality of pupae and adults. The results indicate that there is indeed a distortion caused by a nutritional unbalance. Fitness in females is strongly affected, resulting in a higher proportion of males. During an outbreak, a lower number of females leads to greater competition between males, stimulates the spread in places where resources are available and control the size of the population.

D23SY02PS0689

WHY PARASITIZE ONLY A THIRD OF THE AVAILABLE HOSTS?

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There is strong competition among female *Hyposoter horticola* parasitoids foraging for egg clusters of their host butterfly, *Melitaea cinxia*, in Åland, Finland. Yet only a single wasp gains access to a given host egg cluster, and that wasp only parasitizes about a third of the hosts in the cluster. This behavior appears to be suboptimal for the individual wasp that has many mature eggs in her ovaries, and limited access to future hosts. There are several possible explanations for evolution or maintenance of sub maximal resource use in this system: a) Avoidance of resource over-exploitation through group selection; b) Resource management, which may occur if host individuals in highly parasitized egg clusters perform poorly; c) Risk spreading, such that parasitoids may benefit from distributing their progeny among host egg clusters; (d) Competition with diminishing returns, in which an individual can parasitize more eggs in total by moving from one cluster to another; (e) Density-dependent hyperparasitoid. I will discuss the plausibility of each of these models using empirical data and mathematical modeling, illustrating that some explanations, such as group selection are less plausible than others, such as optimization in response to density dependent hyperparasitism.

D23SY02PS0691

ASSESSING CARIBOU ECOTYPES USING FUNCTIONAL GENES

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Woodland caribou in Ontario use one of two distinct predator avoidance strategies during calving, commonly referred to as behavioural ecotypes. Migratory caribou undergo seasonal shifts in their range from boreal to tundra coasts while sedentary caribou disperse throughout the boreal forest. This project will assess the degree of genetic differentiation between caribou ecotypes within Ontario using functional gene markers. Range overlap and a mixture of caribou lineages in Ontario has made genetic differentiation challenging to detect with traditional neutral markers. Functional genes, however, are subject to selective pressure and may parallel differences between ecotypes. Six genes have been selected for analysis: Mc1R, DRD1, Cyt b, Opsin, IGF1, and PRKG1 several of which are known to vary among caribou subspecies. DNA will be extracted from multiple sources including blood, tissue, and feces. Functional genetic markers will be isolated using Reverse Transcription Polymerase Chain Reaction (RT-PCR) then sequenced using a next-generation sequencer to identify individual genotypes. Once sequenced, a statistical analysis will investigate whether a functional genotype can be ascribed more commonly to a particular ecotype. Determining the relationship between genotype and ecotype can provide insight into the adaptive potential of caribou in response to further northern development. Providing an alternative to mitochondrial classification may help to establish distinctions between caribou ecotypes for management purposes.

D23SY02PS0760

THE ROLE OF POLLINATORS ON DISASSORTATIVE MATING IN NON-RECIPROCAL STYLE MORPHS OF NARCISSUS PAPYRACEUS

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Heterostylous plant populations usually undergo high level of disassortative mating due to differentiated pollen transfer and heteromorphic incompatibility system. Stylar dimorphism lacks perfect reciprocity between morphs, thus it is considered unstable. However, it is very frequent in *Narcissus*. We hypothesize that enough level of disassortative pollen transfer might account for its maintenance. We investigated mating patterns in style dimorphic Narcissus papyraceus, a species with dimorphic and monomorphic populations for the long-styled morph. We set twelve experimental populations in two different sites within the dimorphic and the monomorphic regions and exposed them to natural pollinators. Two different approaches based on paternity analysis revealed increased rates of disassortative mating in most of experimental populations. In a second experiment, we assessed the role of different types of floral visitors on pollen transfer. We set emasculated (receptive) and intact (donor) flowers of each morph in the field, and collected single-visited receptive flowers to examine the pollen load deposited on stigmas. Long-tongued pollinators enhanced disassortative pollen transfer to the short-styled morph, in agreement with paternity experiment. In contrast, short-tongued insects were low efficient pollinators of the long-styled morph, and incapable to pollinate the short-styled morph. Our study provides empirical support for the hypothesis that disassortative mating maintains stylar dimorphism in the genus *Narcissus*. Despite of our first approach could not explain the loss of the short-styled morph in the north range of distribution of the species, the second experiment points out to a possible role of short-tongued insects, as previous correlational studies had proposed.

D23SY02PS0778

SIGNALS OF A DIVERGENT EVOLUTIONARY HISTORY IN THE VOMERONASAL RECEPTOR GENES OF TWO SYMPATRIC MOUSE LEMUR SPECIES

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Most terrestrial mammals possess functional vomeronasal receptors that are used in intraspecific communication and predator recognition. The two types of vomeronasal receptors, V1Rs and V2Rs, bind smaller ligands or larger peptides, respectively. Among primates, mouse lemurs possess the largest V1R repertoire and it has evolved under pervasive and ongoing positive selection. The aim of this study is to analyse and compare the evolution, genetic diversity and demographic history of selected vomeronasal receptor genes in two sympatric mouse lemur species (*Microcebus murinus* and *M. ravelobensis*) in northwestern Madagascar. A total of 40 mouse lemurs (20 of each species) from one study site in the Ankarafantsika National Park were sequenced at 15 different V1R loci and the two recently discovered V2R genes. All selected genes had been shown before to be expressed in the vomeronasal organ of *M. murinus*. *M. ravelobensis* showed a higher genetic diversity as expressed by a higher haplotype and nucleotide diversity than *M. murinus*. A comparative analysis of the respective haplotype networks, mismatch distributions and demographic and spatial modelling will be used to reconstruct the demographic history of both mouse lemur populations and to relate those results to possible Pleistocene colonisation scenarios that were previously developed on the basis of non-coding loci. Funded by VolkswagenFoundation.

D23SY02PS0782

DYNAMICS OF A FAST EVOLVING SEX-RATIO SYSTEM IN NATURAL POPULATIONS OF DROSOPHILA

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Meiotic drive elements that distort Mendelian transmission to their own advantage can rapidly spread in natural populations and leave strong signatures of positive, but not adaptive, selection on the genomes. Among the X-linked segregation distorters known in Drosophila, the Paris system of Drosophila simulans is exceptionally young and can provide important insights into the early dynamics of these elements that kill Y-bearing sperm and cause sex-ratio bias. We studied the biogeography of the Paris driver in Africa and Middle-East and showed that the driving X chromosomes most probably share a recent common ancestor. However, the driver seems to show different local dynamics, probably related to the population structure of D. simulans. In particular, contrary to the Indian Ocean where they are declining, the driving chromosomes seem to have been dramatically rising in the Middle-East during the last decade with a consistent signature of selective sweep. These results provide one of the first exemples of drive suppression strengthening as the driver frequency increases and shows that such selfish elements and the resulting arms race can evolve very rapidly in natural populations.

D23SY02PS0791

ASSESSING THE EFFECTS OF HUMAN SELECTION THROUGH FISHERY ON SEA CUCUMBER GENETIC STRUCTURE AND POPULATION DYNAMICS

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Increased fishing pressure has resulted in widespread overexploitation of populations, with declines in overall abundance of stocks and average size; adverse genetic selection leading to loss of potential fecundity; reduced average spawning size; change of sex-ratio and interspecific equilibrium; and loss of genetic diversity. After the failure of traditional management measures, marine protected areas have been strongly advocated as an ideal tool for the management of coastal fisheries protecting critical spawning stock biomass, intraspecific genetic diversity, population age structure, recruitment supply and ecosystem balance. The sea cucumber fisheries have increased world wide and several species (*Holothuria polii, H. tubulosa* and *H. mammata*) are now targeted in Turkish waters reaching a total production around 600,000 kg in the last year. In this work, we assess the fishery effects on the genetic structure and population dynamics of three sea cucumbers species. We found significant differences in *H. tubulosa* and *H. polii* among protected (Kusadasi) and non-protected (Ayvalik) localities considering their length and weight, showing Kusadasi the largest and heaviest individuals. *H. mammata* did not shown significant differences. Also, we found higher genetic diversity for the three target species (number of total and exclusive haplotypes, haplotype and nucleotide diversity, polimorphic sites) in Kusadasi (marine protected area) than Ayvalik.

D23SY02PS0807

SPINY HAIR IN WILD RODENTS: A ROLE FOR THE ECTODYSPLASIN A RECEPTOR (EDAR) GENE?

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The amino acid change Val370Ala in the Ectodysplasin A receptor (Edar) is associated with hair differentiation in East Asia human populations. Transgenic experiments in mouse have shown that this mutation enhances Edar signaling in vitro, which in turn alters multiple aspects of its hair morphology including straighter hair fibers, increased diameter and more cylindrical form compared to hair of European or African origin. We studied whether this substitution is responsible for the spiny hair or spines observed in some rodent species, a trait that evolved independently at least six times within this lineage. To characterize variation in wild rodents, we first described structural variation in the dorsal hairs of 111 specimens from eight species that represent six rodent families: Heteromyidae, Cricetidae, Echimyidae, Erethizontidae, Hystricidae and Muridae. We found two major patterns: (i) presence of a dorsal groove associated with longitudinal side ridges of hairs in most lineages, and (ii) a roughly circular hair cross-section, without furrows and ridges, in South and North American porcupines. Thus, high levels of ellipticity observed in the Edar transgenic mouse hair were not observed in the wild rodents included in this study. Instead, two novel mechanisms that convert a guard hair into a spine were identified. Second, we sequenced 320 base pairs of exon 11 in Edar which encompassed the SNP implicated in spiny hair in humans. All 11 species had a Valine residue at site 370. We therefore conclude that the non-synonymous substitution in Edar previously associated with hair morphology does not play a role in modifying wild rodent guard hairs; instead different mutations in Edar and/or other genes are responsible for parallel phenotypes. Together, we demonstrate that different evolutionary pathways can produce similar morphology, in this case spiny hairs in mammals.

D23SY02PS0812

SEXUAL NETWORKS: MEASURING SELECTION IN STRUCTURED POPULATIONS

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Sexual selection is traditionally measured at the population level, assuming that populations lack structure. However, increasing evidence undermines this approach, indicating that intrasexual competition in natural populations often displays complex patterns of spatial and temporal structure. This complexity is due in part to the degree and mechanisms of intra- and intersexual interactions, which influence the intensity and scale of sexual competition. Attempts to measure selection at the local and global scale have been made through multi-level selection approaches. However, definitions of local scale are often based on physical proximity, providing a rather coarse measure of local competition, particularly in polyandrous populations where the local scale of pre- and post-copulatory competition may differ drastically. These limitations can be solved by social network analysis, which allows us to define a unique sexual environment for each member of a population: 'local scale' competition, therefore, becomes an emergent property of a sexual network. Here, we propose a novel quantitative approach to measure pre- and post-copulatory sexual selection, integrating multi-level selection with information on local scale competition derived as an emergent property of networks of sexual interactions. We then use simple simulations to illustrate the ways in which different patterns of pre- and postcopulatory competition can impact estimates of sexual selection. We show that for low to intermediate levels of polyandry, the proposed network-based approach provides substantially more accurate measures of sexual selection than more traditional population-level approaches. Furthermore, our approach highlights how the scale of pre- and postcopulatory competition can differ vastly. We argue that the increasing availability of fine-grained behavioral datasets provides exciting new opportunities to develop network approaches to study sexual selection in complex societies.

D23SY02PS0885

DIVERGENT SELECTION ON, BUT NO GENETIC CONFLICT OVER, FEMALE AND MALE TIMING AND RATE OF REPRODUCTION IN A HUMAN POPULATION

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The sexes often have different phenotypic optima for important life-history traits and because they share much of their genome, this can lead to a conflict over trait expression. In mammals, the obligate costs of reproduction are higher for females, making reproductive timing and rate especially liable to conflict between the sexes. While studies from wild vertebrate populations show support for such sexual conflict, it remains unexplored in humans. We used a pedigreed human population from preindustrial Finland to estimate sexual conflict over age at first and last reproduction, reproductive lifespan and reproductive rate. We found that the phenotypic selection gradients differed between the sexes. For age at first and last reproduction and reproductive lifespan, the relationships with fitness (number of grandchildren) tended to be nonlinear in women, suggesting an intermediate optimum value, while they were linear in men. Both sexes showed a linear decrease in fitness with increasing reproductive rate. We next established significant heritabilities in both sexes for all traits. All traits, except reproductive rate, showed strongly positive intersexual genetic correlations and were strongly genetically correlated with fitness in both sexes. Moreover, the genetic correlations with fitness were almost identical in men and women. For reproductive rate, the intersexual correlation and the correlation with fitness were weaker but again similar between the sexes. These findings illustrate that apparent sexual conflict at the phenotypic level is not necessarily indicative of an underlying genetic conflict and further emphasize the need for incorporating a genetic perspective into studies of human life-history evolution.

D23SY02PS0922

STRONG ADAPTIVE DIFFERENTIATION BETWEEN POPULATIONS OF THE ALPINE PERENNIAL HERB ARABIS ALPINA IN NORTHERN AND SOUTHERN EUROPE

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Reciprocal transplant experiments can be used to determine the magnitude of adaptive differentiation among natural populations and to help identify putative adaptive traits and selective agents. In reciprocal transplant experiments, we quantified local adaptation between Scandinavian and Spanish populations of the alpine perennial herb *Arabis alpina*. At the Scandinavian field site, survival and fruit production of Scandinavian populations were higher than those of Spanish populations, while the opposite was true in Spain. The magnitude of the home advantage varied among years and was highest in a drought year in Spain and after a cold winter in Sweden. The results suggest that differences in tolerance to drought and cold contribute to adaptive differentiation between populations from the two regions. They further suggest that these *A. alpina* populations represent a highly suitable model system for examining the functional and genetic basis of plant adaptation in alpine environments.

D23SY02PS0924

DETECTING AND MEASURING SELECTION ON QUANTITATIVE TRAITS IN A RANGE EXPANSION SCENARIO WITH SPATIALLY-EXPLICIT SIMULATIONS

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For at least 50 years now, evolutionary biologists have been discussing about the relative roles of natural selection and genetic drift in shaping the genetic composition of populations. Range expansions are of particular interest in this discussion: They normally occur over environmental gradients allowing local adaptation to take place, but also the demographic properties of these expansions potentiate genetic drift effects that can generate drastic changes in allele frequencies as the populations spread (i.e. allele surfing). Here, we address the detection and measurement of selection in such scenario using simulations. We mimic a range expansion over a variable selective gradient where individuals have in their genomes both loci that are neutral and loci determining a quantitative trait subject to selection. The responsiveness of different summary statistics to the selective pressure is then assessed, and estimates of the selective pressure are made – based on these statistics – with Approximate Bayesian Computation (ABC). Preliminary results, based on a one-locus/two-allele architecture for the quantitative trait, showed that some statistics, such as the slope of isolation-by-distance (IBD) for the quantitative trait (FSTQ), and the difference of slopes of IBD produced for quantitative and neutral loci (FSTQ-FST) are indeed sensitive to selective pressure. Others are currently being investigated. Even though improvements of this model are required (e.g. more complex genetic architectures), the approach of taking into account the spatial dimension of differentiation in quantitative traits offers a promising avenue for investigating the role of natural selection in range-expansion scenarios. We believe that the system developed here can soon be applied to detect and measure natural selection in wild populations. This will allow us to study their possible adaptations to new environments by taking advantage of the robust ABC framework.

D23SY02PS0942

MACRO-ECOLOGY OF PARENTAL CARE IN ARTHROPODS: ARE THE BENEFITS OF EGG PROTECTION HIGHER IN THE TROPICS?

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Parental care is a fundamental aspect of the reproduction of various species, and its costs and benefits are subjected to the influence of both biotic and abiotic factors. Given that biotic interactions, such as parasitism and predation, are stronger at lower latitudes, the benefits of parental care in term of offspring protection are expected to be higher in the tropics when compared to temperate regions. Here we formally tested this macro-ecological prediction using arthropods as model organisms. We used a meta-analysis of experimental field studies that investigated whether unguarded broods in the tropics suffer higher mortality than unguarded broods in temperate regions due to the higher intensity of predatory activity in lower latitudes. Our sample includes so far 39 species belonging to 10 orders of insects and arachnids. Preliminary results indicate that background brood survival is lower in tropical regions, suggesting higher predation/parasitism intensity. However, we found little evidence that the magnitude of the benefit of parental care is greater in tropical regions than in temperate ones. Interestingly, the proportion of tropical species that exhibit additional forms of brood defense (e.g., protective egg coating and nests) is greater than that of temperate species, further suggesting that predation and parasitism are stronger at lower latitudes. These results shed light on how biotic interactions can shape the maintenance of parental care and may also explain the high frequency of parental care among tropical arthropods.

D23SY02PS0955

THE FITNESS IMPACT OF SEED FECUNDITY IN A LONG-LIVED UNDERSTORY SHRUB: A 12-YEAR STUDY

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The transition between seed and adult stages in plant populations involves seed dispersal, germination and seedling survival the seedling/juvenile stage being frequently the most vulnerable phase. In woody plants, the transition from seedlings to reproductives may be a long lasting one and consequently highly stochastic process, which has been rarely documented in the wild because of practical difficulties. However, only disparity among individuals differing in a certain trait in next-generation reproductive offspring will allow natural selection to occur. In this study, I followed the fate of ca. 2900 sown seeds of the long-lived, evergreen shrub *Daphne laureola*, under natural conditions up to their first reproductive event twelve years after sowing. In summer 2000, single-seeded mature drupes were collected from natural crops of 18 mother plants in Cazorla mountains (SE Spain). Seeds were deprived of the pulp and sown in 41 plots according to a split-plot design. Plots were located in two sites differing in abiotic conditions associated with differences in elevation and canopy composition. Germination was relatively high (ca. 70 %) and occurred primarily in the first spring season after sowing, although delayed germination occurred in ca. 10 %. Mortality during the first growing season was high (ca. 55 %) with almost all dead seedlings having experienced herbivory by invertebrates (slugs and caterpillars) at early development stages. Remarkably, only 13 individuals had reached reproductive stage in 2012 suggesting that conditions for establishment were more stringent than conditions for germination. Here, I will analyse whether germination, short-term (1-2 years) and longterm (11 years) survival assessments provided similar results regarding the effects of mother plant (genetics) and sowing location (environment) in this understory shrub characteristic of well-preserved Mediterranean mountain forests.

D23SY02PS0960

MUTUAL ORNAMENTS IN PIED FLYCATCHERS: GEOGRAPHICAL PATTERNS OF ASSORTATIVE MATING

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In many sexually dichromatic taxa, females express, albeit often to a lower degree, male-like traits. Fitness consequences and signaling content of (mutual) ornamentation are often studied for each sex separately, with the exception of assortative mating studies. Male and female pied flycatchers (*Ficedula hypoleuca*) have a conspicuous white wing patch that varies in size and ultraviolet reflectance. This trait affects intra- and intersexual social interactions and may signal individual quality. Positive assortative mating patterns and their fitness correlates, using wing patch data from several populations across pied flycatchers' breeding range. Preliminary results show that within-pair similarity in wing patch differs between populations. Together with current work on spatial patterns in selection for this trait in each sex, these results suggest that local environmental and ecological conditions may affect mutual ornaments differently in males and females.

D23SY02PS0979

PATTERNS OF CORK OAK (QUERCUS SUBER L.) POPULATION DIFFERENTIATION IN CANDIDATE GENES FOR ADAPTIVE TRAITS

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Cork oak is the most abundant tree in Portugal, which together with its economical and social importance, has granted it the status of National Tree. It is the only species that produces renewable commercial cork, occurring across a vast range of climatic conditions in its natural range of distribution, the Mediterranean region, where it exists for over 60 Ma. So processes of local adaptation are likely to have occurred. Identifying candidate genes underlying genetic differences for adaptive traits can help to understand how species have adapted to their environment and to predict how they will respond to future climatic changes, which is a special concern in regions such as the Mediterranean Basin, where a substantial decrease in precipitation and a pronounced warming is expected in the near future. Here, we intend to assess cork oak potential capacity for adaptation by analysing population differentiation and diversity in candidate genes related with adaptive traits and test for associations with environmental variables. Five candidate genes for bud burst and osmotic stress were analyzed (in total, 1890 bp) in 25 populations from its distribution range. From the 19SNPs analyzed, several showed differences between populations, which might be involved in adaptive responses of cork oak. Our analysis, showed the presence of common haplotypes among various populations but some differences in the frequency of alleles between populations. A correlation study of allele frequency/genotype with phenotypic and environmental conditions, revealed some significant correlations between specific heterozygous genotypes and characteristics such as plant height, altitude and rainfall. We expect that further exploitation of these data will bring some insights on the molecular basis controlling local adaptation. Funded by FCT, grant SFRH/BD/68670/2010, projects PTDC/AGRGPL/104966/2008 and PTDC/AGR-AAM/104364/2008.

D23SY02PS0990

IDENTIFYING SIGNATURES OF NATURAL SELECTION IN CORK OAK (QUERCUS SUBER L.) GENES

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Cork oak (Quercus suber L.) is an evergreen tree species holding a great economical and social relevance within its distribution range, the western Mediterranean Basin, associated with cork production. This tree is exploited mainly in orchards known as montado, which comprise great biodiversity. Despite its importance, cork oak stands have been facing a significant decline and climate changes expected to occur during this century may aggravate this decay. In this scenario, the assessment of adaptive genetic variation is essential to understand how cork oak may cope with these threats and to delineate management strategies of its genetic resources. In this work, we started by validating single nucleotide polymorphisms (SNPs) detected through the analysis of cork oak pyrosequenced transcriptome, for a set of putative functional genes. Six fragments were then selected to be analysed with the purpose of finding signatures of natural selection. Samples from populations representing the species' entire distribution range were sequenced and several neutrality tests were performed as well as environmental association tests. Two gene fragments showed signals of balancing selection: the Arabidopsis thaliana orthologs non-expresser of pathogenesis related 1 (NPR1), involved in plant defence response against pathogens, and *auxin response factor 16 (ARF16)*, a gene previously identified as a candidate gene for drought resistance. Additionally, in a class I heat shock protein (*HSP*), one amino acid position was detected as possibly being under positive selection and associated with several precipitation variables, revealing its potential relevance in adaptation to local climatic

conditions. In this study the first steps were taken to start unveiling important information on cork oak adaptation to biotic and abiotic environmental conditions. Funded by Fundação para a Ciência e Tecnologia (FCT): projects PTDC/AGR-GPL/104966/2008 and EST/036/2009.

D23SY02PS0996

CHARACTERIZING THE ADAPTIVE HISTORY OF INFECTION IN HUMAN CYTOMEGALOVIRUS (HCMV)

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Beginning with Sewall Wright's Shifting-Balance Theory, the field has long debated the relative importance of models of population subdivision with migration in dictating the mode and tempo of adaptation. And yet, analytical results and statistical tests of selection appropriate for selection in structured populations have been lacking. We will present recent theoretical results on sweeps in such non-equilibrium models, which lend themselves to a new class of haplotype based test statistics for identifying and quantifying recent adaptive fixations. We apply these results to time-sampled data of Human Cytomegalovirus – a natural population on which to utilize this model given compelling evidence for not only between but also within host population structure – and evaluate the history of adaptation associated with immune-related mechanisms.

D23SY02PS1009

COMPARATIVE TRANSCRIPTOMICS ANALYSIS TO IDENTIFY CANDIDATE GENES FOR A PIGMENT TRAIT IN A CICHLID FISH

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One of the most fascinating questions in evolutionary biology is to integrate morphology, behavior and genetics to uncover the mechanisms of speciation and adaptation. The pigment on male anal fin of cichlid fish plays a key role in breeding behavior, providing an ideal model to study morphology-behavior-genetics interactions in adaptive radiation. Here, we make use of comparative transcriptomics analysis to generate candidate genes involved in the formation of a pigment trait on the male anal fin of a cichlid fish from Lake Tanganyika, which will be useful for further studies on the emergence and evolution of this fascinating novel trait.

D23SY02PS1040

GENETIC BASIS OF LOCAL AND TEMPORAL ADAPTATION IN NATURAL DAPHNIA POPULATIONS

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Ongoing global warming may cause long-lasting changes in local environmental conditions. If the established populations fail to adapt to these changes, they will possibly be forced to migrate or face extinction and become displaced by external, preadapted populations. We studied natural populations of the water flea *Daphnia galeata* to get insights into the genetic changes associated with the rapidly changing conditions in their habitats. We used resting eggs of *D. galeata* obtained from multiple layers of sediment cores along a latitudinal transect across Europe. This enabled us to not only compare populations across their geographic range but also over an extended time period. Presumably temperature relevant candidate genes *ERNA*, *TRY5F* and *MHC1* were sequenced and 11 putatively neutral microsatellite loci were analyzed in every population. Signals of local adaptation were detected using a comparison of genetic differentiation at functional candidate and neutral microsatellite loci and outlier tests. Mantel tests showed a correlation between genetic differentiation within candidate gene *TRY5F* and temperature differences between habitats. These results suggest local adaptation in *Daphnia* as a response to various temperature regimes. Future studies will link field observations (natural selection at temperature relevant genes) with genotypic architecture of clonal lineages (exposed to different temperature regimes) and gene expression analyses.

D23SY02PS1110

BOYS AND GIRLS AND PLANT-EATERS: IS HERBIVORY SEX-BIASED IN MERCURIALIS PERENNIS?

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Sex-biased herbivory has been implicated as one of the driving forces in the evolution of dioecy. Different partitioning of resource sources and sinks in male and female plants allows different defense strategies. Along with constitutive and induced defences, plants can tolerate herbivore damage to a certain extent, and can ameliorate the pressure through changes in phenology and life history. Genderdimorphic species of genus Mercurialis are informative model systems in addressing questions in plant evolutionary ecology, including interactions with herbivores. For Mercurialis annua, the most commonly studied species of the genus, herbivore prefference for male plants has been demonstrated in experimental conditions. The aim of our study was to explore the pattern of herbivore damage in natural populations of *Mercurialis perennis*, a dioecious perennial herb with wide geographic and altitudinal distribution. Plants were sampled from a range of habitats in Serbia. Damage to leaf area was scored and used as an estimate of herbivore load. Herbivore damage was examined in relation to plant gender, altitude, plant height and reproductive allocation. Our results do not confirm the pattern of male-biased herbivory in natural conditions. The variance of herbivore damage between the sexes among the habitats was large, with highly significant effect of site, and non significant effects of sex and sex-site interaction. In a montane population, herbivore damage was male-biased, but without statistical significance. Studied populations showed male-biased sexual size dimorphism. The study of *Mercurialis perennis* offers a promising approach to better understanding the effect of plant gender on general vigor, tolerance and resistance to herbivores.

D23SY02PS1155

ANT PREADAPTATION TO THE URBANIZED TERRITORIES: CASE STUDY

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Urbanization is now one of the greatest threat caused on wildlife by human activity. Its negative impact consists of habitat and community destruction, high level of chemical, thermal and light pollution etc. Urban entomofauna is greatly depleted, but extinction is not random. Recently it has been shown that black garden ant *Lasius niger* (Hymenoptera: Formicidae) is the most robust to the urbanization pressure. It becomes even more abundant in the most disturbed areas in Moscow city. This species is also very interesting for ecological and evolutionary researches because it is widespread, in contrast to the genetically most studied ant species. We sequenced *L. niger* genome using NGS technology to search for possible adaptation to the urbanization pressure. Several results which can be treated as preadaptation were revealed. For example, detoxification system is often involved in the urban adaptation. We found that this species has an expansion of cytochrome P450 gene subfamily 9 which is unusual to hymenopterans – this subfamily is expanded in humans. This feature could possibly play a role in preadaptation to the urbanized environments. The next stage of the study is the search for the possible selective sweeps in urban populations.

D23SY02PS1236

ADAPTIVE TE INSERTIONS IN DROSOPHILA: NATS, MIRNAS, AND PIRNAS

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Recent Transposable Element (TE) insertions in *Drosophila melanogaster* are a useful tool to identify adaptive mutations. The objective of this study is moving from the identification of a putatively adaptive TE insertion to the molecular mechanisms and associated fitness effects. We have focused on a TE insertion located in the 3'UTR region of *Kmn1* gene. The 3'UTR of *Kmn1* overlaps with the 3 'UTR of its nearby gene, *CG11699*, giving rise to a cis-natural antisense pair of transcripts (cis-NATs). We found that besides being incorporated into the 3'UTR of *Kmn1*, the TE insertion also affects the transcript length of *CG11699*. Specifically, the TE disrupts the GU-rich downstream element of the distal polyA signal of *CG11699* and as a consequence a shorter transcript is produced. These structural changes are very likely to have functional consequences since the presence of the TE adds (*Kmn1*) and eliminates (*CG11699*) miRNA binding sites, introduces piRNA binding sites (*Kmn1*) and affects the length of the overlapping region between these two genes. Indeed, we detected that this TE insertion is associated with an under-expression of *Kmn1* and an over-expression of *CG11699*. In order to identify the phenotypic effect of this insertion, we first focused on *CG11699* since this gene is involved in the activation of *Aldh-III*, an enzyme involved in xenobiotic metabolism. We found that the TE insertion is associated with an increased survival rate after an acute exposure to benzaldehyde, a prototypical drug classically used to assess Aldh-III activity, strongly suggesting that this TE confers resistance to xenobiotic stress. Our results show that a single mutational event has broad molecular consequences that can be translated into ecologically relevant phenotypic effects. This example reinforces the idea that TEs are a powerful natural tool for genome evolution.

D23SY02PS1301

SKIN-MUCUS BACTERIA COMMUNITIES IN COTTUS (PISCES) SPECIES INHABITING SECONDARY CONTACT ZONES

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The composition of microbial communities associated with plants and animals may depend on the environment as well as on host genetics. However, it is unclear to what extent host microbiota interactions represent an evolutionary determinant that drives adaptation to different habitats. A prerequisite to study such questions is to disentangle whether host microbiota associations are caused by genetic or environmental factors. Natural hybrid zones can contain genetically diverse individuals that are exposed to varying environmental factors. This study aims at testing whether a fish hybrid zone contains sufficient host – microbiota – environment associations for in depth studies. We analyzed the microbial composition of the dermal mucus in two species of freshwater *Cottus* inhabiting a secondary contact zone in the River Sieg system near Bonn, Germany. For this purpose genomic DNA was extracted from fin samples from wild-caught individuals that were collected in different habitats. The samples were genotyped using *Cottus* specific SNP markers to determine the genetic background of each individual. Bacteria specific 16S rRNA markers were amplified and next-gen sequenced to study the bacteria associated with each fish. We explored microbial diversity and abundance based on database searches. Our results suggest that different species of Cottus are associated with specific bacterial taxa and that the bacterial composition in *Cottus* hybrids depends on host's genetic factors as well as the environment. This paves the way to identify the host genomic regions underlying variation in bacterial communities as well as to experimentally study the effects of environmental factors.

D23SY02PS1392

SIGNATURES OF DIVERGENCE IN PERIPATRIC POPULATIONS OF THE INVASIVE ANT CARDIOCONDYLA OBSCURIOR

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Worldwide small propagules of terrestrial and aquatic species are constantly transported to novel habitats through human activity. This global experiment on peripatric speciation enables us to evaluate the role of random genetic drift and effective population size in the formation of isolated populations with distinct phenotypes. Here we describe alternative social phenotypes of two introduced populations of a highly inbred ant. We compared the two populations for signatures of divergence using a whole-genome SNP based approach. In addition we tested for differences in aggression behavior within and between populations and analyzed variation of cuticular chemical profiles. Our data support the idea that effective population size can play a role in determining social phenotype in ants.

D23SY02PS1419

TROPICAL AFRICAN CATTLE A UNIQUE MODEL FOR THE STUDY OF LIVESTOCK ADAPTATION

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Indigenous cattle populations of Africa have a unique history which makes them particularly suitable for the understanding, at the genome level, of environmental adaptation. African taurine cattle Bos taurus taurus were the first to arrive on the continent. They were followed by the humped Bos taurus indicus zebu from their centre of domestication on the Indian subcontinent. The result of these migrations is a mosaic of zebu-taurine cattle population adapted to a diversity of local environments (e.g. semi-dry desert, humid and sub-humid forested areas) and production systems (e.g. pastoral, small-holder). Here, we use genomic tools to unravel genetic positive signature of selection in the East African shorthorn zebu, a locally indigenous adapted cattle from Western Kenya. We show that its autosomal genome structure is a complex make-up of taurine and zebu background while its mitochondrial DNA genome is of taurine origin only. Positive signatures of selection are detected through high density SNP chip genotyping and full genome sequencing. They include genes which may be associated with adaptation to disease challenge or reproductive function. An example of these genes is RXFP2, which is involved in testicular descent development, and horn development. Analysis of the mitochondrial DNA reveal signature of purifying selection against deleterious mutations.

D23SY02PS1458

NAVIGATION SYSTEM AND SPATIAL MEMORY OF A SOLITARY WASP

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The specialist wasp *Hyposoter horticola* is a parasitoid of the Glanville fritillary butterfly (*Melitaea cinxia*), which lives as a typical metapopulation in a fragmented landscape in the Åland Islands in Finland. Adult females parasitize the larvae of *M. cinxia* in the short time period while the host is still in the egg shortly before hatching. There is strong competition among female wasps for host egg clusters. The foraging female wasps learn the location of host egg clusters and monitor them until they become susceptible to parasitism. This is a very time- and energy-consuming process, therefore a huge evolutionary investment. It is not yet known how *H. horticola*, or any insect other than social bees and ants, navigates through the landscape, which navigational system is used, and what spatial learning occurs. Ants, social bees and nesting bees have a nest as a fixed starting point for navigational context. In my research I study which navigational system is used by *H. horticola*, a cognitive map or path integration in order to compare this to closely related social insects. I am using an array of RFID systems positioned at host egg clusters to track the foraging path of wasps equipped with laser-activated RFID chips. I am presenting the results of the research of my first field season.

D23SY02PS1459

GENOMICS OF INTROGRESSION ACROSS A SPECIES BARRIER: HALF A MILLION SNPS ACROSS THE EUROPEAN HOUSE MOUSE HYBRID ZONE

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Understanding speciation as a birth-death process requires study not only of the maintenance of species barriers, but also of those forces acting to break them down ('speciation genes' and 'anti-speciation' genes). We use the European House Mouse hybrid zone as a natural experiment to examine such forces. When SNP marker data is dense on the recombination map it cannot be treated as a series of independent loci but can, instead and more naturally, be analysed in terms of Fisher's representation of the outcome of admixture and recombination: the ancestral source of blocks of DNA along the genome and the position of junctions (arising from recombination) where that source state changes. Cascades in the size of blocks allow ancestral polymorphism to be distinguished from introgression polymorphism. Frequency spectra of introgressed block sizes allow inference of the time since secondary contact. Geographic outlier analysis of blocks allow regions of the genome associated with the introgression of 'anti-speciation' genes across the species barrier to be identified. These insights increase our understanding not only of the mouse species barrier, but also how species barriers can decay in their natural settings.

Symposium

3. Molecular Evolutionary Innovations

23 and 24 August



Program

Friday 23 August Session(s): 12, 13

Saturday 24 August

Session(s): 14

Organisers: Frédéric Brunet and Rita Ponce

Invited speakers: Vaishali Katju and Dan I. Andersson

Description:

Evolutionary innovations, including evolution of new functions, bear the hallmarks of genome plasticity. During this symposium we will consider studies that have discovered and traced-down these events, mechanisms involved in their origin, the dynamics and fate of novelties, and the tools allowing their retrieval at the genomic level. Emphasis will be given to studies detailing changes at the molecular and cellular level up to those showing behavioural impact.

D23SY03IT15:45R2

COPY-NUMBER CHANGES IN EXPERIMENTAL EVOLUTION: RATES, FITNESS EFFECTS AND ADAPTIVE SIGNIFICANCE

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Gene copy-number differences due to gene duplications and deletions are rampant in natural populations and play a crucial role in the evolution of genome complexity. The rate at which new gene copies appear in populations greatly influences their evolutionary dynamics and standing gene copynumber variation in populations. The duplication rate may therefore have profound effects on the role of adaptation in the evolution of duplicated genes with important consequences for the evolutionary potential of species. In this talk, I will discuss three long-term experimental evolution experiments in *Caenorhabditis elegans* that we have utilized to investigate fundamental properties of the gene duplication process. First, we conducted oligonucleotide array comparative genome hybridization (oaCGH) on *C. elegans* mutation accumulation (MA) lines subjected repeatedly to single-worm bottlenecks each generation to provide the first direct estimate of the spontaneous genome-wide rate of duplication in a multicellular eukaryote. The gene duplication rate in *C. elegans* is quite high and exceeds the spontaneous rate of point mutation per nucleotide site in this species by two orders of magnitude. Second, I discuss new oaCGH results of low-fitness experimental lines subjected to adaptive recovery via population expansion to investigate whether copy-number variants (CNVs) constitute a common mechanism of adaptive genetic change during compensatory evolution. Lastly, long-term spontaneous MA lines maintained at three varying effective population sizes for >400 generations were used to investigate whether CNVs accumulate differentially under varying intensities of natural selection and provide some insights into their average fitness effects.

D23SY03IT17:45R2

EVOLUTION OF NEW GENES BY GENE AMPLIFICATION AND DIVERGENCE

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New genes can evolve or be acquired in at least three different ways: (i) from a pre-existing gene copy via gene amplification and subsequent divergence of the extra copyies to acquire a new function., (ii) de novo from short open reading frames located in non-genic DNA sequences and (iii) by lateral gene transfer via transformation, transduction and conjugation. With regard to the amplification-divergence mechanism, Ohno's dilemma presents a central problem. That is, how can a duplication be selectively maintained and at the same time free to mutationally diverge and acquire a new function? One solution to Ohno's dilemma is the Innovation-Amplification-Divergence (IAD) model that suggests that evolution of new genes occurs under continuous selection for maintenance of the original copy and divergence of the extra copy. Experimental tests and sequence-based data support the IAD model as one mechanism for the evolution of new genes.

D23SY03RT16:33R2

ORIGINS AND FUNCTIONAL EVOLUTION OF LONG NON-CODING RNAS IN TETRAPODS

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The search for molecular evolutionary innovations strongly relies on comparative analyses of gene repertoires, sequences and expression patterns. Such studies have been instrumental for our understanding of the genetic basis of lineage-specific phenotypes and of individual gene functions. However, these analyses have so far been restricted to protein-coding genes, and the contribution of other categories of genes, such as long noncoding RNAs (lncRNAs), has yet to be explored. Here, we use RNA sequencing to identify lncRNAs in eleven tetrapod species and we present the first large-scale evolutionary study of lncRNA repertoires and expression patterns. We identify ~11,000 primatespecific lncRNA families, which show evidence for selective constraint during recent evolution. These "young" lncRNA genes are lowly transcribed and predominantly expressed in testes, in agreement with the hypothesis that the permissive chromatin state of the testis favors the emergence of new genes. Interestingly, we also identify ~2,400 highly conserved lncRNAs, including ~400 genes that likely originated more than 300 million years ago. We find that lncRNAs, in particular ancient ones, are generally actively regulated and may predominantly function in embryonic development. Most lncRNAs evolve rapidly in terms of sequence and expression levels, but global patterns like tissue specificities are often conserved. We compared expression patterns of homologous lncRNA and protein-coding families across tetrapods to reconstruct an evolutionarily conserved co-expression network. This network, which surprisingly contains many lncRNA hubs, suggests potential functions for lncRNAs in fundamental processes like spermatogenesis or synaptic transmission, but also in more specific mechanisms such as placenta growth suppression through miRNA production.

D23SY03RT16:57R2

HORIZONTAL GENE TRANSFER IS AN INNOVATIVE MECHANISM IN ASEXUAL EUKARYOTES TO ACQUIRE NOVEL FUNCTIONS

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Asexuality is considered an evolutionary dead-end for metazoans, but bdelloid rotifers challenge this view as they are assumed to have persisted asexually for millions of years. Neither male sex organs nor meiosis have ever been observed in these microscopic animals. Females form oocytes through two successive mitotic divisions, with no reduction of chromosome number and no indication of chromosome pairing. In addition, bdelloids are able to survive extreme desiccation at any stage in their life cycle. Supporting long-term absence of meiosis and sexual reproduction, the genome architecture of the bdelloid rotifer Adineta vaga presents a high level of shuffling and rearrangement incompatible with conventional meiosis. Using an alien index approach, 8% of protein-coding genes in the A. vaga genome are probably of non-metazoan origin. Confirming this abundance of genes of non-metazoan origin, it has been estimated that at least 8% of the genes in *Adineta ricciae*, a related bdelloid species, are of foreign origin. This suggests that horizontal gene transfers (HGT) happen in bdelloids far more often than in other eukaryotes studied so far. For instance, this is twice the proportion observed in rootknot nematodes (3-4% of foreign genes) that were considered as holding the record for metazoan animals. In these nematodes, HGT have been shown to play an important role in the emergence of plant parasitism. In *A. vaga*, gene families involved in resistance to oxidation and carbohydrate metabolism, essential for desiccation resistance, are significantly expanded and many have been acquired through HGT. Quantitative PCR studies confirm the expression of foreign genes related to desiccation resistance in the bdelloid species A. vaga. Whether acquisition of foreign genes via HGT is a key innovative mechanism in eukaryotes allowing adaptation to changing environments in the absence of sexual reproduction is an important question that deserves further investigation.

D23SY03RT18:33R2

MOLECULAR TINKERING OF NANOMACHINES: THE EVOLUTION OF A DIRECT PROTEIN DELIVERY SYSTEM FROM THE BACTERIAL FLAGELLUM

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Protein secretion systems drive bacterial virulence, symbiosis and competition. Some components are homologous between systems and/or with other cellular appendages. This suggests extensive evolutionary tinkering of the molecular components of secretion systems and other complex cellular membrane machineries. Here, we present a scenario for the evolution of the non-flagellar type 3 secretion system (NF-T3SS). This system allows the direct injection of proteic effectors into eukaryotic cells, and is involved in both beneficial (symbiosis) and antagonist (pathogenicity) relationships between bacteria and eukaryotes. This system is partly homologous to the bacterial flagellum. We developed tools to detect NF-T3SS in genomes, and used phylogenomics and comparative genomics to study the evolution of this system. We could show that NF-T3SS derived from a flagellar ancestor in a series of steps, each representing accretions to the system of proteins from other molecular machines. This resulted in a secretion system that diversified and adapted to different types of hosts and ecological associations. We also found a new intermediate conserved system in myxococcales, which emerged from the ancestral NF-T3SS and is neither a direct protein delivery system nor a flagellum. Thus secretion systems have intricate histories. This study and ongoing work should help deciphering the common evolutionary history of different secretion systems. It also provides clues on patterns of evolution of the large molecular structures that drive ecological interactions.

D23SY03RT18:57R2

EVOLUTIONARY INNOVATIONS IN SEX DETERMINATION MECHANISMS DRIVEN BY WOLBACHIA BACTERIAL ENDOSYMBIONTS IN THE ISOPOD ARMADILLIDIUM VULGARE

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In the isopod Armadillidium vulgare, genetic sex determination follows female heterogamety (ZZ males and ZW females). However, many A. vulgare populations harbor maternally-inherited Wolbachia bacterial endosymbionts which can convert genetic males into phenotypic females, leading to populations with female-biased sex ratios (1). The W sex chromosome has been lost in lines infected by Wolbachia and all individuals are ZZ genetic males. The female sex is determined by the inheritance of Wolbachia by the A. vulgare individual, thereby leading to a shift from genetic to cytoplasmic sex determination. We are using comparative genomics and expression profiles to identify Wolbachia gene(s) responsible for feminization of A. vulgare males. Surprisingly, some A. vulgare lines exhibit female-biased sex ratios despite the lack of Wolbachia. In these lines, female individuals are ZZ genetic males carrying an unknown feminizing factor. To elucidate the genetic basis of female sex determination in these lines, we sequenced the genome of a female by illumina. We identified a large piece of the Wolbachia genome transferred to the A. vulgare nuclear genome. The transferred genomic fragment shows non-Mendelian inheritance and co-segregates perfectly with the female sex in pedigrees, in agreement with observed biased sex ratios. These results suggest that sex determination in these A. vulgare lines is under the control of nuclear gene(s) of bacterial origin. Overall, our results indicate that Wolbachia bacteria can drive shifts in sex determination mechanisms in A. vulgare. More generally, they emphasize that bacterial endosymbionts can be powerful sources of evolutionary novelty for fundamental biological processes in eukaryotes, such as sex determination. This research is funded by an ERC Starting Grant (EndoSexDet) to RC. (1) Cordaux et al. (2011) The impact of endosymbionts on the evolution of host sex-determination mechanisms. Trends in Genetics. 27, 332-341.

D24SY03RT10:30R8

AY-LIKE SOCIAL CHROMOSOME CAUSES ALTERNATIVE COLONY ORGANIZATION IN FIRE ANTS

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Intraspecific variability in social organization is common, yet the underlying causes are rarely known. In the fire ant Solenopsis invicta, the existence of two divergent forms of social organization is under the control of a single Mendelian genomic element marked by two variants of an odorant-binding protein gene4–8. Here we characterize the genomic region responsible for this important social polymorphism, and show that it is part of a pair of hetero- morphic chromosomes that have many of the key properties of sex chromosomes. The two variants, hereafter referred to as the social B and social b (SB and Sb) chromosomes, are characterized by a large region of approximately 13 megabases (55% of the chromosome) in which recombination is completely suppressed between SB and Sb. Recombination seems to occur normally between the SB chromo- somes but not between Sb chromosomes because Sb/Sb individuals are non-viable. Genomic comparisons revealed limited differenti- ation between SB and Sb, and the vast majority of the 616 genes identified in the nonrecombining region are present in the two variants. The lack of recombination over more than half of the two heteromorphic social chromosomes can be explained by at least one large inversion of around 9 megabases, and this absence of recombination has led to the accumulation of deleterious mutations including repetitive elements in the non-recombining region of Sb compared with the homologous region of SB. Importantly, most of the genes with demonstrated expression differences between individuals of the two social forms reside in the non-recombining region. These findings highlight how genomic rearrangements can maintain divergent adaptive social phenotypes involving many genes acting together by locally limiting recombination.

D24SY03RT10:54R8

THE RAPID LIFE CYCLE OF DROSOPHILA ORPHANS

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Orphans are genes restricted to a single phylogenetic lineage. They emerge at high rates and frequently provide a selective advantage or even essential functions to their host. While these features predict an accumulation of genes, the gene number has remained remarably constant through evolution. This paradox of a stable gene number in the presence of a high rate of gene birth has not been resolved. Because orphan genes were mainly analyzed over large evolutionary time scales, orphan loss, a key factor of orphan evolution, remained unexplored. Here, we study the patterns of orphan turnover among close relatives in the *Drosophila obscura* group. We show that orphans are not only emerging at a high rate, but they are also rapidly lost. The pattern of orphan loss is clearly non-random: young orphans are more likely to be lost than orphans, which originated earlier. Furthermore, highly expressed orphan genes with a strong male-bias are more likely to be retained. Since lost and retained orphans show similar evolutionary rates, but lineage specific functional requirements.

D24SY03RT11:18R8

UNSTABLE TANDEM REPEATS IN PRIMATE PROMOTERS ENHANCE EXPRESSION DIVERGENCE

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Tandem repeats in eukaryotic gene promoters can change gene expression drastically due to their extremely low stability. We hypothesized that unstable tandem repeats in promoters increase expression divergence along the primate phylogeny. A search for tandem repeats in promoter regions of 13,000 human, chimpanzee and macaque orthologous genes revealed that 30 % of primate promoters contain tandem repeats. Genes driven by these repeat-containing promoters show significantly higher rates of expression divergence. More specifically, we found a significant correlation between repeat instability and expression divergence. This relation might explain gene expression divergence in more special cases, as in gene duplication events or in disease formation.

D24SY03RT11:42R8

THE PARADIGMA OF IRREVERSIBILITY IN GAMETOPHYTIC SELF-INCOMPATIBILITY: THE MALUS AND PRUNUS SYSTEMS

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In the present hypothesis, S-RNase-based gametophytic self-incompatibility (GSI) evolved once before the split of the Asteridae and Rosidae, but the Rosaceae Malus and Prunus GSI systems are different: in Prunus the S-RNase gene presents two introns and the S-pollen is a single F-box gene (called SFB). The two genes show evidence for a partially coevolved history, both presenting high levels of synonymous and non-synonymous divergence, as well as positively selected amino acid sites that account for the specificities present in natural populations. In Malus, the S-RNase gene has one intron, and multiple S-pollen F-box genes (called SFBBs) have been described. Levels of diversity at the S-RNase gene are 10 times higher than at the SFBB genes. Nevertheless, intra-haplotypic diversity of SFBB is similar to the S-RNase gene. Moreover, there is evidence for amino acids under positive selection only when intra-haplotype SFBB genes are analyzed. Thus, it is not surprising that different self-recognition mechanisms have been proposed: in Prunus self S-RNases are protected from degradation by the self-SFB protein, while in Malus each SFBB protein is predicted to interact with a subset of non-self S-RNases that mediates their degradation. Here, we perform a detailed characterization of S-RNase and F-box like genes present in the P. persica and M. domestica genomes. Phylogenetic analyses revealed three duplications that predate Malus and Pyrus speciation. The gene lineage determining GSI function are different in Malus and Prunus. Expression analyses in 12 Malus tissues revealed that two of the S-RNase lineage genes are expressed mostly in pistils, but one has acquired seed expression. In Malus the closest SFB-like gene shows the highest expression in stamen but is also expressed in seeds. An hypothesis for the evolution of Malus and Prunus GSI is presented. Implications of the polyphyletic S-RNase GSI evolution on uncharacterized GSI systems are discussed.

POSTERS

D23SY03PS0018

GENOMIC STRUCTURES AND GENE EVOLUTION ON MAMMALIAN X CHROMOSOMES

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Of genomic structures, intrachromosomal segmental duplications (ISDs) are relatively large repeats in neighboring regions on a chromosome. Counting of ISDs on each human chromosome found more ISDs on X than any of the other chromosome. In this study, we show that the concentration of ISDs are not a general characteristic for X chromosomes, or not due to sequence-specificity of the human X chromosome, but are correlated with gene evolution and expression control. ISDs on the X chromosome in four mammalian species with different origins or evolutionary histories (human, mouse, opossum, and platypus) were identified and characterized. The number or the size of ISDs was different among these species; ISDs on human and mouse X chromosomes were much larger in size, in the number, and more structurally complex than those in opossums. Moreover, gene density and the number of different gene families in ISDs-containing regions were larger in the human and mouse, whereas in the opossum, ISD-regions were gene-poor. Interestingly multiple X chromosomes of platypus did not have any ISDs, except for one on the X1 chromosome. These observations indicated that ISDs accumulated on the X chromosome in the eutherian ancestor. In the human, more than 70% of the genes within X chromosomal ISDs were cancer-testis antigen (CTA) genes, and they are highly expressed in testis and cancer cells. The CTAs showed primate or eutherian-lineage specific, suggesting the recent origin and rapid evolution within ISDs. The amplification and complexity of ISDs can be evolutionally maintained by the emergence and functional constraint of CTAs, respectively. In addition, the accumulation of X chromosomal ISDs might be involved in the mechanism for expression control, because the ISD-regions on X chromosomes were low-methylated and genes within the ISDs were expressed specifically in germ cells.

GENE DUPLICATIONS: A ROLE IN ADAPTIVE EVOLUTION

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Evolutionary potential is limited by the number and type of genes present, but how these limits shape the evolution of new functions remains a matter of debate. In this context, gene duplications are thought to be the main source of raw material for new evolutionary features: duplications are essentially envisioned as genomic substrates for long-term adaptation. Their early evolution is thus often neglected, notably how gene-dosage modifications affect fitness and condition their first steps. The evolution of insecticide resistance in the mosquito *Culex pipiens* is one of the few examples of contemporary duplications. Resistance represents a genetic adaptation to the environmental changes induced by insecticides, and as such, provides evolutionary biologists with a contemporary model for studying parameters that influence ongoing adaptation. The *ace-1* gene encodes the acetylcholinesterase (AChE1), the target of organophosphate (OP) insecticides. A mutation in this gene causes high resistance levels in many mosquito species. However, a strong genetic constraint drives resistance evolution, as the degree of resistance and the ability to degrade ACh trade off. Recently, we identified in *Cx. pipiens* new *ace-1* alleles that carry one susceptible and one resistant copy associated on the same chromosome. These different duplicated alleles show different dynamics in the field. We propose that duplications are selected to disentangle the two functions, i.e., by improving synapse signaling and mosquito's fitness while maintaining resistance. I will present our recent work investigating 1) the duplication origin at the molecular level and 2) the complex gene-dosage/fitness relations and their impact in the field dynamics of these innovations. Our work stresses the role of duplications as immediate adaptive features, but shows that their fate is checked by natural selection early on: only those passing its short-term sieve can become seeds for future evolution.

MULTIFUNCTIONAL VITELLOGENIN – CAN THE EVOLUTION OF A PLEIOTROPIC GENE BE LINKED TO THE DOMAIN ARCHITECTURE OF ITS PROTEIN PRODUCT?

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How do single proteins perform a broad range of tasks, and how can evolution act on pleiotropic genes/proteins? Vitellogenin is a multitask protein mostly associated with its egg-yolk function. It transports lipids to the egg and provides a source of amino acids for the embryo, and this function is conserved across several taxa. However, in the honey bee workers, vitellogenin is also a central lifehistory regulator that supports immune cell viability, protects against oxidative stress, and suppresses risky foraging behavior. It is expressed in queens, workers and males. In fish, this egg-yolk protein has bactericidal effects and is overexpressed in infected females and males. The key to this multifunctionality may lie in the evolution and diversity of specific structural domains. Typical structural elements of vitellogenins are, among others, N-sheet, alpha-helical and the vertebrate phosvitin domain. The latter has been pinpointed as the bactericidal actor in fish. In the honeybee, it is known that different vitellogenin domains are evolving under differing selection pressure, the putative receptor-binding N-sheet being more conserved than the other parts of the sequence. Furthermore, honeybee vitellogenin is a structurally adjustable protein that can shed the N-sheet domain. We have identified the alpha-helical domain as a membrane-binding region. Using the methods of cell and molecular biology, we show that the membrane-binding can be linked to vitellogenin's antioxidative and putative anti-inflammatory functions in the honey bee. Thus, research on protein domains can facilitate understanding the evolution of pleiotropic genes such as *vitellogenin*.

3. Molecular Evolutionary Innovations

D23SY03PS0297

HOTWIRING THE FLAGELLUM: RAPID, REPEATABLE REWIRING OF A GENE REGULATORY NETWORK IN *PSEUDOMONAS FLUORESCENS*

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Pseudomonas fluorescens, a common microbial inhabitant of the soil and plant commensal, uses flagella for swimming motility and a biosurfactant known as viscosin for sliding motility – both are required to maximise colonisation efficiency in the plant environment. The master regulator gene that differentially regulates flagellum expression is FleQ. Both $\Delta fleQ$ mutants and those carrying functional mutations in the viscosin synthase genes maintain partial motility in liquid agar (0.25%) using the other function as a motility mechanism. However, loss of function in both the viscosin synthase and FleQ genes completely abolishes *in vitro* surface motility. Serendipitously, we observed a SBW25*fleQ*-viscoin mutant regained motility over agar surfaces after about 72 hours of starvation on agar plates. This heritable motility phenotype was observed with a range of $\Delta fleQ$ mutants with independent mutations in viscosin genes following starvation selection. Electron microscopy showed motile evolved mutants were able to express flagella. Further growth analysis revealed major growth defects in minimal medium, and genome sequencing revealed the bacterium had "rewired" its genetic circuitry, co-opting homologous genes from the nitrogen regulation pathway, resulting in restored expression of the flagella at the cost of reduced efficiency in the nitrogen cycle. This work highlights how bacteria can use simple innovations to adapt to potentially catastrophic genome mutations.

3. Molecular Evolutionary Innovations

D23SY03PS0394

HOW IMPORTANT ARE SEX CHROMOSOMES IN SEXUAL DIMORPHISM?

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There is an extensive body of evolutionary theory predicting that the sex chromosomes should play a large role in encoding sexually dimorphic phenotypes. Although gene expression often follows these theoretical predictions, phenotypic studies have been far less conclusive. Many studies have focused on a gene-by-gene analysis of expression differences between males and females, and although this approach has yielded many new insights, it can fall short in explaining how an expression change in one sex-linked gene can affect the expression pattern of other autosomal genes. To better understand the complex regulatory structure underpinning sexual dimorphism, we employ a network-based approach based on gene co-expression in the chicken, Gallus gallus. We used RNA-seq data from males and females in several tissues and across development to construct a global gene co-expression network. Genes on the Z chromosome are more connected to male-biased autosomal genes. Additionally, male-biased Z-linked genes have a significantly higher connectivity in comparison to unbiased Z-linked genes. Finally, we detected sex-biased sub-networks, revealing that sex-biased genes on the sex-chromosomes are tightly connected with sex-biased genes on the autosomes. Taken together, these analyses suggest that the Z chromosome plays a large regulatory role in male-biased expression, and therefore male-specific phenotypes. Our analysis also shows the potential of network approaches that integrate large-scale transcriptomic datasets in a systems biology framework for revealing the influence of the sex chromosomes on general sex-specific gene expression patterns.

ON THE FAST TRACK TO EXTINCTION? ORIGIN, DIVERGENCE AND MUTATION RATES OF ASEXUAL ORGANISMS

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Recent research explored the parameters under which sexual reproduction is favourable. However, it is not clear whish genomic background allows for asexual reproduction to be established and which mechanisms confine parthenogenetic species to terminal phylogenetic branches with evolutionary short lifespans. One genetic condition observed in asexual species is polyploidy arising through hybridisation. The possession of multiple copies per gene could temporarily aid in buffering against detrimental mutations that cannot be eliminated by outcrossing in asexuals and thus accrue in genomes. Still, losing the 'best' genotype forever once it has mutated, asexual organisms might be trapped in a feedback loop culminating in extinction - especially as low quality genotypes might evolve elevated mutation rates. Conversely, elevated mutation rates could help in adaptation to novel and extreme environments where asexuals are often found. Employing a mutation accumulation (MA) experiment I have explored the mutation rate in a parthenogenetic species of the cosmopolitan nematode genus Panagrolaimus, analysing a closely related hermaphroditic species in comparison. I sequenced and assembled the genomes of both species in order to construct references against which genomic sequences of MA lines were mapped. I then differentially screened for mutations and directly calculated rates. To analyse the origin and genetic background of parthenogenesis in *Panagrolaimus* I also generated genomic and transcriptomic data of two dioecious and several parthenogenetic species. Here I am presenting results from these analyses, giving a first estimate of the mutation rates and divergence. My genomic and cellular data indicate that asexual *Panagrolaimus* species are tetraploid hybrids. This study will aid in a better understanding of evolutionary forces acting on asexual organisms and is a basis for research on molecular and cellular mechanisms leading to the establishment of parthenogenesis.

EVOLUTIONARY "TINKERING" IN THE ORIGIN OF THE INSECT TERMINAL PATTERNING SYSTEM

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A key early process in development is the determination of the embryonic axes. The anterior-posterior axis in insects is determined by a series of signaling pathways and transcription factors. These are best known from the fruitfly *Drosophila melanogaster*, where the torso pathway activates a number of posterior transcription factors, while interacting diffusible factors define the anterior. We have cloned the homologues of most of the key players in terminal patterning from the milkweed bug *Oncopeltus fasciatus*, focusing on *huckebein, torsolike, hunchback, orthodenticle* and *tailless*. We then studied their expression and function, and their interaction with other early developmental pathways. Our results show that many of the pathways known to be involved in *Drosophila* are derived from the more ancestral roles still preserved in *Oncopeltus*. We use our results to discuss a model for the evolution of the terminal patterning system in insects, and show that the evolution of this pathway is a classic example of evolutionary "tinkering", where different elements are co-opted independently into a single novel patterning system.

3. Molecular Evolutionary Innovations

D23SY03PS0546

GENETIC ARCHITECTURE PROMOTES THE EVOLUTION AND MAINTENANCE OF COOPERATION: THE EVOLUTIONARY CONSTRAINT OF CODING OVERLAPS ON FUNCTIONALLY UNRELATED GENES

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When cooperation has a direct cost and an indirect benefit, a selfish behavior is more likely to be selected for than an altruistic one. Kin and group selection do provide evolutionary explanations for the existence of stable populations of cooperators in nature, but we still lack the full understanding of the genomic mechanisms that can prevent cheater invasion. We used Aevol, an agent-based, in silico genomic platform to evolve populations of digital organisms that compete, reproduce, and cooperate by secreting a costly public good. We found that populations of phenotypically equal cooperating individuals often have very different abilities to resist cheater invasion. To understand the underlying mechanisms, we performed bio-inspired genomics analysis by determining and comparing the locations of metabolic and secretion genes, as well as the relevant promoters and terminators. We found that populations of cooperators characterized by the strong association between metabolic and secretion genes (promoter sharing, overlap via frame shift or sense-antisense encoding) were more robust to cheater invasion than ones where such association was weak. We performed mutation analysis of the evolved individuals and determined that the accessibility of mutations decreasing cooperation without decreasing overall fitness was negatively correlated with the amount of operons and overlap between secretion and metabolism. Effectively, cooperation evolved to be protected and robust to mutations through overlapping genetic architecture, especially when cooperation was costly. Due to operon sharing and gene overlap, even when mutations that eliminate cooperation appear, they are likely to be selected against due to their simultaneous and direct negative effect on fitness. Our results uncover an important genetic mechanism for the evolution and maintenance of cooperation, and suggest promising methods for preventing loss of genes introduced into biological synthetic organisms.

PROTEIN DOMAIN EMERGENCE AND ARRANGEMENTS BEAR HIGH POTENTIAL FOR RAPID EVOLUTIONARY INNOVATIONS AND ENVIRONMENTAL ADAPTATION

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True novelty is only rare in nature. Most biological systems use core units (or modules) which are rearranged and reused in various contexts. Here, we explore protein domains – the modules of protein evolution, structure and function - and how domain rearrangements can create diversity and evolutionary innovation in only a few steps and surprisingly fast. We study the dynamics of domain rearrangements for genomes of both plant and insect species. We demonstrate that protein domains are frequently reused to create new arrangements and functionality. By using a phylogenomic approach, we can explain the formation of around 80% of all new arrangements by simple modular events such as fusion, fission or terminal domain loss in just one step. The remaining new arrangements frequently consist of many repeat domains occurring in complex patterns and seem to result from multi-step events. Whereas many domains are lost along every lineage, newly emerging domains are found to be often evolved in stress response and developmental innovations. They appear to spread swiftly and to be associated with environmental adaptation. While we identify relatively large and well-conserved core sets of single-domain proteins, long multi-domain arrangements tend to be species-specific indicating their importance for functional novelties. We find that modular rearrangements of protein domains have impact on all levels of the cellular signaling apparatus and subsequently may have strong adaptive potential. In summary, we investigate the high volatility of single domains and complex arrangements. We highlight nature's potential to make use of protein domains for rapid evolutionary and functional innovation. The work has been carried out in collaboration with the following colleagues: E. Bornberg-Bauer, A. K. Huylmans, A. R. Kersting, A. D. Moore, and A. Schüler. Moore et al (2013), Biochim Biophys Acta. doi: 10.1016/j.bbapap.2013.01.007. Kersting et al (2012), Genome Biol Evol. 4(3):346-59.

GENOMIC CHANGES DURING THE EVOLUTION OF INCREASING SPECIALIZATION IN FUNGUS-FARMING ANTS

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Ants are an ecologically important group of insects with a vast array of specialized lifestyles and symbioses. One of the most spectacular ant symbioses is fungus farming, found in a single clade of New World (attine) ants, a mutualism to which the ants contribute plant material in exchange for food provided by the fungal crops. Since this mutualism evolved in the Amazon basin from hunter-gatherer like ancestors some 50 mya, it has undergone a series of evolutionary transitions of which the use of specialized rather than generalist fungal strains, active herbivory rather than using dead plant parts, polymorphic rather than monomorphic worker castes, and multiple rather than single mating of queens are the most important. The advent of high-throughput sequencing techniques now allows these questions to also be addressed at the molecular evolutionary level. In a recent study, we showed that the genome sequence of the leafcutter ant Acromyrmex echinatior, a representative of the most highly derived leaf-cutting fungus-farming ants, has characteristic changes in detoxification pathways, loss of function in arginine metabolism pathways, and expansion of specific peptidase gene-families relative to other ant genomes. We have now sequenced and partly analyzed the genomes of five additional fungusfarming ant species, representing all phylogenetic branches of the higher attine ants and a lower attine outgroup, and thus most of the major evolutionary transitions. These comparative genomic analyses allow us to assess rates of gains and losses of genes and variation in lineage-specific selective pressures, differences that we hope to connect to some of the phenotypic adaptations in the respective lineages.

PRIMARY AND SECONDARY STRUCTURES OF GROUP II INTRON NADI GENE IN PETOTA SPECIES

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Group II introns are self-splicing, mobile elements, which share a common ancestry with the eukaryotic spliceosome and had fundamentally influenced on organization of terrestrial genomes. Here, we report primary and secondary structures of group II intron of mitochondrial Nad1 gene (b/c intron) in 12 Solanum (sect. Petota) species. b/c Nad1 intron length varied from 1513 bp in S. tuberosum to 1483 bp in other 11 potato species and tomato S. lycopersicum. The intron length in Nicotiana species taken as outgroup was 1212 bp. The boundaries of all six canonical domains of intron pre-RNA were revealed. Consensus sequences GUGCG at the 5'-and AY at the 3'-end were present in all introns. EBS1-IBS1\EBS2-IBS2 and other motives essential for interdomain interactions were identified. 20 variable sites and three extended indels were detected. 240-bp, 30-bp insertions and 10 SNPs were specific to all Solanum mitotypes. Only 6 SNPs were variable in analyzed Solanum intron sequences characterizing S. gourlayi, S. vernei, S. spegazzinii mitotypes. Most of mutations were localized in IV, I, II domains. V and VI domain sequences were completely invariable. Despite the fact that in Solanum species NadI b\c intron was conservative, some deviations of intron pre-RNA secondary structure in comparison with canonical bacterial introns II were detected. Besides the loss of the maturase ORF in the domain IV, several subdomains like IB, IC2, hairpin ID(ii) have not been detected in the Solanum NadI b\c intron. The domain VI lacked a bulged adenosine at the expected position (7 nucleotides from the 3'-end of the intron) required for the transesterification step of the intron splicing. Instead of it a short tight helix VI with single bulged adenosine at the 10 nucleotides from the 3'-end of the intron was present. These structure deviations can point out quite different evolutionary pathways in group II introns of flowering plants. The work was supported by RFBR grant 11-04-00573.

THE EXTENT OF HORIZONTAL GENE TRANSFER AND ITS ROLE IN THE EVOLUTION OF BACTERIAL SPECIES

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Horizontal gene transfer (HGT) is a key evolutionary force shaping the genomic repertoires of bacterial lineages and playing a major role in bacterial adaptation. In turn, adaptive shifts into new niche habitats may drive other evolutionary processes, such as speciation. Thus, to understand the evolution of bacterial species it is important to elucidate the extent to which HGT contributes to adaptation. Pseudomonas syringae is a plant-pathogenic bacterial species complex comprised of many hostspecific pathovars. The diversification of lineages within this species appears to have involved numerous adaptive shifts onto new hosts. Here, we apply a comparative genomics approach to quantitatively assess the extent of HGT that occurred during the diversification of 27 strains of P. syringae. We have developed an improved analysis pipeline that is able to account for common errors associated with draft genome sequences. For example, we estimated that ~77% of the initial lineagespecific genes were accountable to error. We have determined the core and flexible genome components and used these data to reconstruct the history of gene gain and loss within a phylogenetic framework. We estimated the relationships among these strains using 2,141 core protein alignments, and mapped more than 11,000 gene gain events and 16,000 gene losses across this phylogeny using a maximum likelihood method. Our analyses suggest an extensive recent history of gene gain and loss in *P. syringae*, such that pathovars acquire on average approximately 925 genes within the timeframe of 1% protein sequence divergence within the core proteome. Thus, HGT is the predominant evolutionary force in these lineages, potentially explaining the rapid diversification of *P. syringae* via the differential adaptation of individual pathovars onto different host species. In light of these observations, we revisit the concept of HGT as a facilitator of, rather than a barrier to, bacterial speciation.

3. Molecular Evolutionary Innovations

D23SY03PS1105

CIS-REGULATORY EVOLUTION AND FUNCTIONAL DIVERSIFICATION OF GENE DUPLICATES IN DIPTERA

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Gene duplication plays a major role in evolution of novel gene functions as it provides a material basis for variation and selection. We are interested in elucidating how *cis*-regulatory changes contribute to functional diversification ensuing gene duplication. To address this question we are studying the Three-Finger-Domain Protein/Ly6 gene family in insects. Members of this family encode different GPIanchored membrane proteins and are fully conserved across drosophilids. Our analyses of the sequenced insect genomes indicate that a subset of these genes is unique to higher dipterans. We are focusing our attention on seven paralogues of Drosophila, which our phylogenetic analysis showed to derive from sequential duplications of a single orthologue. In order to determine how their expression domains diversified, we have characterized the embryonic expression patterns of the Drosophila paralogues and their unduplicated orthologues in other insects representing different phylogenetic positions and stages of duplication (the Mediterranean fruit fly, Ceratitis capitata, the scuttle fly, Megaselia abdita, the mosquito, Anopheles, the butterfly, Bicyclus anynana and the red flour beetle, Tribolium castaneum). We found that the original expression domain of the unduplicated orthologue localized predominantly to the developing nervous system, which, upon subsequent duplications, expanded to a wide array of tissues. While a subset of the duplicates retained the tissue-specificity of the unduplicated orthologues, the others acquired novel tissue-specific expression suggesting neofunctionalization. We are currently identifying the *cis*-regulatory elements of the duplicates and the unduplicated orthologues to elucidate the *cis*-regulatory mechanisms underlying the evolution of divergent expression patterns.

ARE THERE PATTERNS IN THE FORMATION AND EXPRESSION OF NOVEL GENES?

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Observed differences between species are often due to differences at genomic level. In particular, the analysis of newly evolved genes provides information that helps us to understand the evolution of novel functions. Here, we examine the mechanisms involved in the formation of novel genes (less than 50 MYA) and we characterize the expression patterns of these genes. This illustrates the variety of mechanisms implicated in the formation of new genes, as well as the preferential location of expression of novel functions. We further analyze the *Sdic* gene family and use it to investigate in greater detail the formation and expression of novel genes. The Sdic gene family, a young gene family in Drosophila *melanoqaster*, represents an interesting window into the evolution of novel genes, as it evolved after the split of *D. melanoqaster* and *D. simulans* i.e. less than 2 million years ago. This gene family originated from a formation of a chimera of two unrelated genes, that was followed by several duplication events, being therefore a candidate for the study of two important mechanisms of gene formation: gene shuffling and duplication. This family is composed by several tandem copies of the gene Sdic, and at least one copy (sdic1) is expressed in testis, incorporated into the sperm tail and encodes a sperm specific intermediate chain. It has recently been showed that although this novel gene is not essential for the development of spermatids and *Sdic* knock-out males are fertile with no effect on the size or sex ratio of their progeny, *Sdic* has a role on sperm competition. This effect of *Sdic* in improving sperm competition suggests that *Sdic* may have had an important role, contributing to the species barrier between *D. melanogaster* and *D. simulans*. The analysis of particular newly evolved genes provides information in finer detail, that we hope can be generalized and will help understand the evolution of new genes and new functions in the genomes.

DETECTION OF SELECTION IN THE GENETIC NETWORKS INVOLVED IN THE SIGNALING OF THYMOPOIESIS

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The emergence of adaptive immune defense has enabled vertebrates to eliminate or prevent pathogen growth. Adaptive immunity is the ability to recognize and remember specific pathogens allowing stronger responses each time the pathogen is encountered. T-lymphoid cells are central players in this event. The adaptive immune systems arouse in basal vertebrates, in the lineage of jawless vertebrates and then further evolved in jawed vertebrates with fundamental differences in the mechanism of recombination and the molecular nature of receptor structure. Although the phylogenetic positions occupied by jawless and cartilaginous fishes (representatives of the most basal groups of extant vertebrates mentioned earlier) are in route of the evolution of the adaptive immune system, the emergence of the process of T-cell development (thymopoiesis), is still uncertain. In order to provide insight to the evolution of such a complex system, we selected a set of 39 genes related with the thymopoiesis, in which the cluster with the highest enrichment score, identified the following functional annotations, hematopoietic or lymphoid organ development, immune system development, hematopoisis and leukocyte differentiation, according to (DAVID; http://david.abcc.ncifcrf.gov), they represent 11 classes, the majority of which are Chordates, 40 are Mammals (7 belong to the order Primates), 8 Actinopterygii, 3 Aves, etc. To quantify the enrichment of slowly and rapidly evolving genes we used a Markov Clustering Algorithm. Phylogenetic analysis revealed that those closer linked to the immune system had higher values of ω then other genes. In average values of ω for each lineage were estimated revealing witch lineages had higher ω values and thus allowing the creation of a matrix of positive Darwinian selection genes. Mutation hotspots were identified in 3D representations of their respective proteins.

HALF OF THE SOX GENES REMAINED DUPLICATED SINCE THE TELEOST SPECIFIC WHOLE GENOME DUPLICATION

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Two successive events of whole genome duplications (WGD) occurred at the base of the vertebrate lineage, coined 1R and 2R for Rounds of WGD. An additional third round of WGD (3R) occurred at the base of the teleostean fish. Sox genes encode a family of transcription factors that has experienced a phase of expansion leading to 20 sox genes well described in human and mouse. This gene expansion preceeded the vertebrates lineage and enhanced even more through both the 1R+2R WGDs and tandem duplication occurrences. In fish, additional sox genes have been described with orthologous relationship assessed by phylogenetical analyses. We were interested to know how have evolved such a group of genes since the 3R event. To this end, we carried out a bioinformatic analysis, searching exhaustively the public releases of fish genomes as well as other public databases. We combined both the phylogenetic information and synteny analyses to assess the evolutionary history of fish sox genes. We found evidence that in fish, 10 of the 20 mammalian orthologues of these sox gene family come from and remain duplicated since their 3R origination. This 50% ratio is way above the estimated global average of 12% of genes that remained duplicated since the 3R event, a value in favor of the idea that transcription factors have played a key role in the diversification of the teleost lineage. In addition, we performed expression analyses of these sox genes in three fishes (zebrafish, medaka and platyfish) and observed species specific expression for some of them, in agreement with this hypothesis.

THE PANCREATIC LIPASE-COLIPASE COMPLEX: AN UNEXPECTED OLD DUO

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Colipase (CPLS) is a small protein cofactor essential for the activation of pancreatic lipase (PL) an enzyme involved in hydrolysis of dietary triglycerides. CPLS gene orthologues have been described exclusively in mammals and birds, in contrast to PL which has a wider phylogenetic distribution. Here we investigate the evolutionary history of the CPLS gene family in metazoans. Our findings indicate that CPLS-like genes first emerged in chordate ancestry. In agreement, we find clear orthologues in the invertebrate chordate species, *B. floridae* and *C. intestinalis*. Similarly, agnathans and chondrichthyans have clear single copy CPLS genes. In contrast, the majority of teleost species examined here lack a CPLS gene. Sequence and structural analysis using comparative homology modeling demonstrated that all CPLSs recovered exhibit a true CPLS profile with conserved cysteine residues involved in the formation of the disulphide bridges and the three finger structural topology. Protein-protein docking analysis demonstrates that the lamprey and spotted gar PL-CPLS complexes show a very similar binding pattern when compared to the crystal structure of H. sapiens. Overall our data indicates that this enzymatic system is older than expected and represents a chordate innovation possibly linked with the emergence of the pancreas.

EVOLUTIONARY STUDY OF RDH10 GENE FAMILY REVEALS A NOVEL MEMBER RETAINED IN ECTOTHERMIC VERTEBRATES

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Nuclear receptor-mediated retinoic acid signalling is crucial for organ modelling and maintenance. Active retinoid biosynthesis involves a two-step oxidation cascade: retinol oxidation followed by retinaldehyde dehydrogenase activity, suggested to coordinate retinoic acid supply. Recent studies have highlighted the functional role of a membrane-associated retinol dehydrogenase (RDH10) in the first oxidation step in mammals. This enzyme displays tissue and time-specific expression patterns that correlate with both retinoic acid and retinaldehyde dehydrogenase activities: suggestive of an additional checkpoint for retinoic acid regulation. Here we investigated the evolution of chordate *rdh10*. While a single copy, *rdh10a*, is observed in birds and mammals, reptiles, amphibians and teleosts exhibit an additional uncharacterized gene, *rdh10b*. Both rdh10a and rdh10b have duplicate copies in teleosts. Phylogenetic and paralogy analysis revealed that vertebrate *rdh10a* and *rdh10b* resulted from whole genome duplication in stem vertebrate evolution; interestingly, with significant functional divergence amongst paralogues. Following duplication, *rdh10b* was lost in warm-blooded lineages and retained in cold-blooded animals. Both enzymes exhibit conserved reaction cores and tridimensional folding; yet, the membrane-association designs appear different: unlike RDH10A, topology predictions advocate for RDH10B solubility. Also, a unique negatively charged insertion is observed in RDH10A isoforms. Gene expression patterns in teleosts, D. rerio and O. nicotilus, were also examined. While *rdh10a* is ubiquitously expressed, *rdh10b* expression is typically restricted to gonads, skin, and brain, and is concomitant with the onset of pigmentation and circulation during teleost embryonic development. Together these results support a functional specialization within the *rdh10* family and emphasize a dichotomy among vertebrates according to thermal homeostasis mechanisms.

Symposium

4. Evolution of Symbiotic Interactions in Communities: Novel Approaches

23 and 24 August



Program

Friday 23 August

Session(s): 11, 12, 13

Saturday 24 August

Session(s): 14

Organisers: Kayla King, Mario X. Ruiz-González, Jérôme Orivel and

Justyna Wolinska

Invited speakers: Christoph Vorburger and Jacobus Jan Boomsma

Description:

Biological interactions within communities shape the evolution of individual species. These relationships (obligate/facultative, beneficial/detrimental) force the emergence of innovative traits. The most exciting research on these topics now combines comparative "omics" approaches with concepts from evolutionary and behavioural ecology. The symposium will highlight recent progress in this interdisciplinary field and will discuss the interplay between communities and evolutionary biology: how do species interactions within a community affect evolution?

D23SY04IT14:00R7

USING MOLECULAR AND 'OMICS' APPROACHES TO UNDERSTAND THE EVOLUTIONARY COMPLEXITIES OF ANT FUNGUS-FARMING

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The attine fungus-growing ants have become true model systems for the study of symbiotic adaptations now that high-quality reference genomes are increasingly available. This has given remarkable opportunities for pursuing molecular approaches to make connections between phenotypic traits and the expression of specific genes. High throughput sequencing techniques are now also allowing exploration of the diversity of additional symbiont communities, although the reconstruction of their function is challenging when they cannot be cultured. I will give an overview of recent developments and identify likely directions of future work.

D24SY04IT10:30R7

SYMBIONT-MEDIATED COEVOLUTION BETWEEN APHID HOSTS AND PARASITOIDS

Christoph Vorburger¹

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The present understanding of host-parasite coevolution is based on models that assume a direct genetic interaction between host and parasite. This assumption has been challenged by an increasing number of observations showing that hosts may rely on 'helpers' in the form of microbial endosymbionts for defence against parasites. Among the best-studied cases are aphids, in which much of the variation in resistance to parasitoids is due to facultative endosymbiotic bacteria. Using this example I illustrate how defensive endosymbionts can alter the reciprocal selection between hosts and parasitoids and thus mediate their coevolution. Heritable symbionts increase the variation available to selection, they make the host-parasite interaction more specific as a consequence of strong genotype-by-genotype interactions between the parasitoids and the aphids' symbionts, and they induce trade-offs, because protection by symbionts does not come for free to the host. Infected aphids suffer from a reduced lifespan and lifetime reproduction in the absence of parasitoids, and the magnitude of this costs is determined by genotype-by-genotype interactions between hosts and their symbionts. I show that this complex three-way interaction can lead to interesting genetic dynamics in simulations that assume some potential for lateral transfer of defensive symbionts, and I show empirically that this potential is indeed realised in aphid-parasitoid systems. Endosymbionts thus play a decisive role for the evolution of their hosts as well as for the evolution of their hosts' natural enemies.

D23SY04RT14:48R7

SERRATIA SYMBIOTICA FROM APHIS FABAE: A STEP FORWARD IN UNDERSTANDING THE GENOMIC EVOLUTION OF A LIFESTYLE TRANSITION FROM FREE-LIVING TO APHID-DEPENDENT

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Bacterial intracellular symbiosis is widespread in insects and exhibits a large variety of phenotypes, ranging from parasitism to mutualism. Aphids developed mutualistic relationships with different levels of dependency, from obligate to facultative endosymbiosis, and with various effects (e.g., metabolic complementation, increased resistance to parasites and thermal stress). Serratia symbiotica was described as a secondary endosymbiont found in many aphid families and playing a role in heat tolerance and protection against parasitoids. We argue that this bacterium represents a good model for studying the evolution of the aphid-endosymbiont relationship parallel with a lifestyle transition of this microorganism, from free-living to aphid-dependent. *S. symbiotica* can be a primary-like endosymbiont in the aphid *Cinara cedri* and a facultative endosymbiont in *Acyrtosiphon pisum* and *Aphis fabae*. The S. symbiotica strain found in A. fabae can also exist in free-living style, cultivated on artificial rich medium. In the present study the genome of the previously isolated strain of S. symbiotica CWBI 2.3 was sequenced and compared to the genome sequences of the uncultivable strains from A. pisum (strain Tucson) and C. cedri. The genome of S. symbiotica CWBI 2.3 was sequenced using Illumina and PacBio sequencing, resulting in a genome size of ~3.5 Mb, which is larger than both previously sequenced genomes of *S. symbiotica* but smaller than the genome sizes of free living *Serratia* species. The genome of S. symbiotica CWBI 2.3 is phylogenetically close to that of the strain Tucson, but it also presents some features that are more related to free-living bacteria, such as the capacity of synthesizing most of the amino acids. The functional and synteny analyses performed so far suggest that the genome of *S. symbiotica* CWBI 2.3 represents a relevant step in understanding the evolution of a lifestyle transition towards dependency on another organism.

D23SY04RT15:12R7

NO HOST IS AN ISLAND: THE IMPACT OF MULTIPLE SYMBIONTS ON PAIRWISE INTERACTIONS

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Host – symbiont interactions, though often studied as a pairwise process, rarely occur in isolation. In most cases individual host populations often interact, and potentially coevolve, with multiple symbiont populations concurrently. These pairwise coevolutionary interactions will be affected by the biotic environment in which they take place and vice versa, however the complexity of natural populations make these effects hard to unravel in the wild. Experimental evolution provides an excellent framework in which to examine the impact of these rival symbiotic interactions, as (co)evolutionary dynamics can be observed through time in communities of known biotic structure. Using this approach we examine the conflict between two otherwise independent symbionts of the same host; a parasitic lytic phage and a facultatively mutualistic plasmid, both native to the bacteria *Pseudomonas fluorescens*. We find that the presence of a second symbiont has major effects on the ecology and evolution of these interactions in both directions. Firstly we show that the presence of coevolving phages accelerates the loss of plasmids under conditions in which they are parasitic, but not mutualistic. Secondly we demonstrate that plasmid carriage alters the trajectory of bacteria-phage coevolution, favouring generalist phageresistance strategies among hosts also carrying the plasmid. This study therefore shows the impact community complexity has on pairwise host – symbiont interactions, and in turn how these interactions can shape the community in which they are embedded.

D23SY04RT15:45R7

COMPARATIVE GENOMICS OF WOLBACHIA AND THE BACTERIAL SPECIES CONCEPT

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Bacteria display a wide diversity of specialized interactions with insects. In the intracellular niche, bacteria evolve under selection for host-interaction processes, at the same time as being members of what may be a complex community of other endosymbionts. Among the obligate endosymbionts, Wolbachia have an unparalleled host range infecting at least 20% of all insect species as well as other invertebrates. The Wolbachia are divided into supergroups, where supergroup A and B strains are the most common in insects. Interestingly, multiple infections with strains of both supergroups are common, but how they interact with each other and the host is largely unknown. Evolutionary studies of endosymbiont communities are challenging, due to the fastidious nature of these bacteria. We have developed a novel method, based on multiple-displacement amplification (MDA), which has allowed us to sequence and assemble two new *Wolbachia* strains co-infecting the fruit fly *Drosophila* simulans, and representing supergroup A and B respectively. A comparative genomics study including additional strains revealed 24 and 33 supergroup-specific genes, putatively involved in host-adaptation processes. We are now investigating the genome expression of *Wolbachia* in *D.simulans* at the proteome level, in order to evaluate the role of the supergroup-specific genes. Recombination frequencies were high for strains of the same supergroup, while the inferred recombination fragments for strains of different supergroups were of short sizes. Surprisingly, the genomes of the co-infecting Wolbachia strains were not more similar to each other and did not share more genes than other A- and B-group strains that infect different hosts. This suggests that the supergroups are irreversibly separated and that barriers other than host-specialization can maintain distinct clades in recombining endosymbiont populations.

D23SY04RT16:09R7

PLANT PERFORMANCE DISPARITIES IN TRIPARTITE MUTUALISTIC ASSOCIATIONS

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Ant-plants are interesting model systems of protective mutualisms in which the associated ants are assimilated as agents of constitutive indirect defense. The plants provide ants with nesting space and food in exchange for indirect benefits such as protection. The outcomes of the relationship might, however, vary according to the behavior of the interacting ant partners, thus differently influencing plant fitness. Here, we explore this framework by studying a two-to-one ant-plant mutualism between two closely-related ant species, Allomerus decemarticulatus and A. octoarticulatus, competing for the same host-plant species, *Hirtella physophora*. In these systems, both ant species specifically cultivate a third partner, an Ascomycota fungus. The fungus plays a structural role that the ants use it in constructing gallery-shaped traps to ambush prey and by penetrating into the plant tissue, the hyphae also enhance plant nutrition in both systems. We also examine how the behavioral ecology of these two ant species affects host-plant fitness. First, we show that differences in the efficacy of the predation strategy (ambushing and patrolling leaves) induce a variation in the intensity of the biotic defense, affecting plant growth and fruit production. Moreover, these two ant species impose reproductive costs on their host-plant by partially destroying floral buds. This castration behavior favors the production of more leaves for the host-plant (nesting space for the ants) thanks to the reallocation of plant energy from reproduction to growth. However, plants totally castrated experimentally produced notably smaller domatia and extrafloral nectaries. Such a decrease in the investment in myrmecophytic traits demonstrates the existence of inducible sanctions against too virulent castrating ants. Altogether, these results show how multiple, co-occurring symbiont species (and their traits) influence host-plant performance by modulating their context-dependent outcomes.

D23SY04RT16:33R7

FACULTATIVE SYMBIONTS AS A EUKARYOTIC HORIZONTAL GENE POOL

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Facultative or 'secondary' bacterial symbionts are very common in eukaryotes, particularly insects. While not essential, they often provide beneficial functions that can profoundly affect host biology. It has been hypothesised that secondary symbionts may form a "horizontal gene pool" shuttling adaptive genes amongst lineages in an analogous manner to plasmids and other mobile genetic elements in bacteria. However, the viability of this hypothesis rests upon a key unknown. We do not know if the distributions of symbionts across host populations reflect random acquisitions followed by maternal inheritance, or if the associations have occurred repeatedly in a manner that is consistent with a dynamic horizontal gene pool from which adaptations can be readily gained and lost in response to environmental changes. Here we test the importance of horizontal transfer using the phylogenetic and ecological distributions of secondary symbionts carried by 1104 pea aphids collected from 14 countries and 11 plant-associated populations. This reveals that not only is horizontal transfer common, it is associated with aphid lineages colonizing new ecological niches, such as switching to novel host plants or moving into new climatic regions. Moreover, aphids that share the same ecologies in different regions worldwide have independently acquired related symbiont genotypes, suggesting a central role of the symbionts in their host's niches. In sum, our data reveals symbiont populations are dynamic with their distributions shaped by horizontal transmission and differential retention amongst aphid lineages, both of which are strongly influenced by ecological factors. We conclude that the secondary symbiont community forms a horizontal gene pool that is central to the adaptation and distribution of their insect hosts

D23SY04RT16:57R7

SPECIFICITY AND STABILITY OF THE ACROMYRMEX-PSEUDONOCARDIA SYMBIOSIS IN CHANGING ENVIRONMENTS

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Fungus-growing ants live in a complex symbiosis involving both fungal and bacterial partners. Among these are Actinobacteria of the genus *Pseudonocardia* that are maintained on the ant cuticle to produce antibiotics, primarily against a parasitic fungus of the garden symbiont. The symbiosis has been assumed to be a hallmark of evolutionary stability, but this notion has been challenged by culturing and sequencing data. We used 454 pyrosequencing of 16S rRNA to estimate the diversity of the cuticular bacterial community of the leaf-cutting ant *Acromyrmex echinatior* from Panama. We used field and lab samples of the same colonies, the latter after colonies had been kept under laboratory conditions for up to 10 years. We show that the bacterial communities are highly colony-specific and stable over time. The majority of colonies (25/26) had a single dominant *Pseudonocardia* strain and only two strains were found in the Gamboa population across 17 years, confirming an earlier study. The microbial community on newly hatched ants consisted almost exclusively of *Pseudonocardia* while other Actinobacteria were identified in lower abundances on older ants. These findings are consistent with recent theory predicting that mixtures of antibiotic-producing bacteria can remain mutualistic when dominated by a single vertically transmitted strain.

D23SY04RT17:45R7

SYMBIONT MEDIATED EXTINCTION: EXAMINING THE SPREAD AND EFFECTS OF A MALE-KILLER

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The ubiquity of reproductive manipulating symbionts in arthropods makes them a key component in the ecology and evolution of natural populations. Yet questions surround the dynamics of symbiont spread within and between host populations, and the extent to which their phenotype influences host populations in ecological time remains enigmatic. Ultimately these two issues are linked, since the severity of the symbiont's effect on its host population will be determined by its prevalence, which is a product of its transmission dynamics. These organisms, which manipulate the reproductive biology of their host to aid their own transmission, have a dramatic impact on key fitness traits of their host such as sex-ratio, reproductive behaviors and fecundity. They are therefore an important selection pressure on the host population and, by terminally reducing the effective population size through a combination of these effects, they have the potential to drive their host to extinction. Here we experimentally investigate the spread, maintenance and loss of a heritable male-killing symbiont under differing transmission regimes. We demonstrate that ecological conditions favouring the spread of the malekilling symbiont, Arsenophonus nasoniae, reduced the effective host population size through maledeath, causing severe bottlenecks and eventually driving experimental wasp populations extinct. If reproductive symbionts can sweep to near fixation in local populations, such microbes can have catastrophic effects on the survival of their hosts in the field. This study highlights the potential importance of other symbionts or sex-ratio distorters in the persistence of natural populations.

D23SY04RT18:09R7

SYMBIOSES CATALYSES NICHE EXPANSION AND DIVERSIFICATION

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Interactions between species are important catalysts of the evolutionary processes that generate the remarkable diversity of life. Symbioses, conspicuous and inherently interesting forms of species interaction, are pervasive throughout the tree of life. However, nearly all studies of the impact of species interactions on diversification have concentrated on competition and predation leaving unclear the importance of symbiotic interaction. Here, I show that, as predicted by evolutionary theories of symbiosis and diversification, multiple origins of a key innovation, symbiosis between gall-inducing insects and fungi, catalysed both expansion in resource use (niche expansion) and diversification. Symbiotic lineages have undergone a more than sevenfold expansion in the range of host-plant taxa they use relative to lineages without such fungal symbionts, as defined by the genetic distance between host plants. Furthermore, symbiotic gall-inducing insects are more than 17 times as diverse as their non-symbiotic relatives. These results demonstrate that the evolution of symbiotic interaction leads to niche expansion, which in turn catalyses diversification.

D23SY04RT18:33R7

THE GUT MICROBIOME OF FUNGUS-GROWING TERMITES EXPRESSES TWO ORDERS OF MAGNITURE MORE DECOMPOSITION GENES THAN THE TERMITES OF THEIR FUNGUS-GARDEN SYMBIONT

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Fungus-growing termites (Macrotermitinae, Blattodea) live in mutualistic symbiosis with a Termitomyces fungus (Tricholomataceae, Basidiomycotina), which aids in the degradation of plant material and is reared on a special substrate (the fungus comb) maintained by the termites through the incorporation of plant material after passage through the termite guts. The association with crop fungi has made this termite sub-family one of the major decomposers in the Old World tropics. It has been suggested that also termite gut microbes may contribute carbohydrate-active enzymes (CAZymes) that aid in the breakdown of plant substrate, but verification of this contention has only recently become technically feasible. We have sequenced a *Macrotermes natalensis* genome (1.3 GB), the associated Termitomyces fungal genome, and a set of caste-specific gut microbiomes of termites to characterize bacterial CAZymes contributions of microbiomes relative to the potential of termites and fungus. We identified 86 termite and 211 Termitomyces CAZyme genes, but found surprisingly high numbers of bacterial CAZyme genes in the worker and soldier gut microbiomes (13903 and 11902, respectively). The CAZymes of bacteria and Termitomyces appear to be complementary, suggesting division of labour between the fungal and bacterial symbionts, with gut bacteria decomposing mostly short chain sugars after Termitomyces has broken down the recalcitrant larger carbohydrates. The gut microbiome of the queen was markedly different and almost completely devoid of plant decomposition enzymes, suggesting a high-nutrient diet consisting of only Termitomyces fungus material. These results shed completely new light on the fungus-growing termite symbiosis, reducing the role of termites to being merely the managers of multicomponent microbial/fungal bioreactor systems in their gardens and guts.

D23SY04RT18:57R7

QUANTIFYING THE WOLBACHIA TURNOVER

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Wolbachia bacteria infect millions of arthropod species, with diverse and extreme consequences, ranging from sex-ratio distortion, mating incompatibilities, to protection against viruses, including vector-born human pathogens. In contrast to other maternally inherited intracellular bacteria such as Buchnera in Aphids, Wolbachia is not engaged in long-term, evolutionary stable associations with its arthropod hosts, as evidenced by the variability of Wolbachia infections between closely related host lineages. Its current distribution is thus best seen as resulting from a dynamic interplay between infection acquisition, through horizontal transfer, and infection loss, possibly fuelled by Wolbachiahost evolutionary conflicts. Here we globally quantify this process by tracking recent acquisition / loss events among 3,600 arthropod lineages spanning 1,100 species from Tahiti and surrounding islands, that is, an estimated 70% of the local biodiversity. Reduced gene flow among islands deepens coalescence time, thus providing a mean to reveal recent changes in Wolbachia infection status. Using DNA barcoding approaches to characterise the host cytoplasmic lineages and their Wolbachia infections, we show that infected specimens within species (that is, differing by no more than 3%) substitutions per site at the mitochondrial locus CO1) have an average 88% chance to share the same infection by descent. This probability sharply drops and reaches 22% when host mtDNA divergence ranges from 3 to 10%, that is, after approximately 3 to 10 million years of divergence. Fitting an epidemiological model to this data, we estimate that uninfected lineages acquire Wolbachia every 2 to 7 million years in average, while infections are lost every 1 to 3 million years.

D24SY04RT11:18R7

INTERACTIONS BETWEEN SPECIES SHAPE PARENTAL INVESTMENT IN THE BURYING BEETLE

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Parental care studies typically focus on intraspecific interactions occurring during reproductive events, such as between mates or between parents and offspring. Interactions between species are often neglected, yet feedbacks between species may shape parental investment. This is particularly important when parents breed on temporary resources for which there is high competition. The burying beetle Nicrophorus vespilloides breeds on small vertebrate carcasses which they bury and cover in antimicrobial exudates. Investment in antimicrobial exudates increases in response to carcass microbial load. Females investing more in antimicrobial exudates seem to protect their current brood at the cost of future reproduction. Beetles also carry phoretic mites that breed alongside them on a carcass. We investigate interactions between beetles, mites and microbes, and how these affect reproductive strategies in the burying beetle. Using breeding experiments we test whether mites have beneficial or detrimental effects on beetles breeding in carcasses presenting different levels of microbial challenge. To gauge the microbial challenges faced by wild beetles, we use metagenomics to assess the microbiome of prepared and unprepared carcasses. We found that females breeding with mites have lower levels of antimicrobial activity, potentially safeguarding future reproduction. However, male lifespan is reduced when breeding with mites, suggesting that interacting with mites is costly for males. Beetles radically change the microbiome of carcasses: prepared carcasses, relative to unprepared, show a shift towards overrepresentation of Gram negative bacteria. In conclusion, characterizing relationships between species in this system unravels ecological pressures shaping parental investment and trade-offs between future and current reproduction. Using a metagenomics approach we are able to identify key players in the bacterial communities which challenge carrion beetles.

D24SY04RT11:42R7

ECOLOGICAL CONSEQUENCES OF DIFFERENT KINDS AND DEGREES OF ANT-BUTTERFLY SYMBIOSES

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Lycaenid butterflies show a wide continuum of symbiotic associations with ants, ranging from no to obligate interactions. Mutualistic caterpillars generally feed their ant symbionts with honeydew from their dorsal nectary organ, and in return receive protection from predators and parasitoids. At the other end of the virulence spectrum are species that exploit ants as inquiline social parasites or brood predators, often drastically reducing the fitness of the host ant colony. Here I will report on a project exploring the population level genetic consequences of this variety of lifestyles among lycaenids, using worldwide sample collections and a range of species. The focus is on how kind and degree of interactions with ants affect dispersal (transmission) and population size of the butterflies. Evolutionary theory suggests that virulent parasitic organisms have higher rates of transmission than mutualistic ones (with negative virulence). At the same time, virulence of the butterflies is expected to be negatively correlated with population size, allowing sustainable recovery of the host ant populations. Finally, the challenge of expanding classical population genetic studies beyond the level of congeneric populations will be addressed.

POSTERS

D23SY04PS0104

HOTSPOTS OF SOCIAL PARASITES OF MYRMICA ANTS

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Colonies of social insects, such as ant nests, represent a well protective environment, which is also a good sources of food. Many organisms have evolved to live within ant societies showing different degrees of interaction, ranging from mutualism to parasitism. Social parasites exploit the colony resources over a long period of time. Myrmica ants are hosts for larvae of lycaenid butterflies belonging to the genus Maculinea as well as for larvae of the syrphid fly Microdon myrmicae. Parasitic larvae spend most of their life cycle inside host nests and, depending on the feeding strategy, they prev on the ant brood or they are fed directly by ant workers. Although it is suppose that social parasites are much rare in comparison to their hosts there are places where a few species of social parasites co-occur exploiting the same pool of resources. The aims of this research were to assess factors influencing the infestation of Myrmica nests, as well as to study the host specificity pattern and parasite distribution in the social parasite community. The study was carried out at two sites inhabited by different social parasite communities, each comprising varying proportions of Maculinea teleius, M. nausithous, M. alcon and Microdon myrmicae. Data were collected twice, at the beginning and in the end of parasite larval development. The most important factor influencing the number of parasitic larvae was the size of ant nests. The biggest Myrmica colonies adopted more larvae and they also provided better conditions, which allow survival of the highest number of parasitic caterpillars. At both communities, competition among social parasites posed strong enough selection on M. teleius population to use Myrmica host species not exploited by other social parasites. Thus, in some cases not only the availability of host species but also the presence of other competitors might shape the coevolution between particular species of social parasites and their ant hosts.

VARIATION IN WOLBACHIA-MEDIATED ANTIVIRAL PROTECTION AND EFFECTS ON VIRAL TITERS ACROSS THE WOLBACHIA PHYLOGENY

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In the last decade, endosymbionts has been shown to play an important role in mediating interspecific interactions. One example is the bacterium Wolbachia, a widespread symbiont in arthropods. Wolbachia infection has been correlated with antiviral protection in different host species, however this conferred phenotype has been proved to be highly dependent on the Wolbachia strain as well as to lead to different outcomes regarding viral titers and possibly transmission of viruses. Indeed, in some cases, protection is associated with resistance, that is a decrease in viral accumulation, whereas in others, viral titer is not affected despite increased survival of the host, suggesting that mechanisms of tolerance are involved. Here we investigated the protective phenotype and associated viral titers in a large range of Wolbachia strains originating from several Drosophila species. One the one hand, this study allows us to assess to what extent protection has contributed to the evolution of both Wolbachia and its host. On the other hand, patterns of resistance/tolerance across the Wolbachia phylogeny should give us some insight about how Wolbachia may affect the evolution and spread of viruses in host populations. Finally, our results will be discussed in the light of available and coming wolbachia genomic data to better understand what genes may be involved in the protection.

D23SY04PS0313

A CRYPTIC CO-EVOLUTION BETWEEN AN OBLIGATORY PATHOGEN AND ITS VECTOR WHEN EACH HAS A DIFFERENT DISTINCT HOST PLANT

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Vector-Pathogen-Plant dynamics are complex due to both direct and indirect interactions between the organisms involved. Key factors for the pathogen dissemination are the survival of its vector and its ability to transmit the pathogen into the plant. A possible conflict may arise between the pathogen and the vector if the pathogen host preference does not coincide with that of its vector. This is the case of the obligatory pathogen- Stolbur type phytoplasma (SP) and its vector- the planthopper Hyalsthes obsoletus; In Europe both share the same preferred host plant whereas in Israel the preferred host of one reduces life history fitness of the other.
br>This conflicting situation calls for a local ecological solution. We found that (1) in Israel SP is not present in the vector's preferred host- Vitex agnus-castus, although the vector feeding on this plant harbors SP. (2) Vines are the preferred and only host of SP in the area. (3) The vector can acquire SP from vines. (4) The vector cannot complete its life cycle on vines. (5) There is no vertical transmission of SP from the vector mother to her offspring. In Israel, unlike in Europe, the vector host plant is not commonly neighboring vineyards. Thus, reaching vines involves a cost to the vector of distance flying and leaving a suitable host for non-suitable plant. Apparently, the pathogen must apply a different strategy in order to disseminate between vines. Previous studies have shown that vector-borne pathogens can alter traits of their host plants and vectors in ways that influence their physiology and behavior. We suggest that the vector is "seduced" by the pathogen to change its behavior by real or assumed benefits. These benefits may be directly induced by the pathogen in the vector or indirect through a change caused by the pathogen to its host plant to attract the vector.

IS YOUR NICHE REALLY YOURS? SYMBIOTIC MICROBES DETERMINE ADAPTATION TO THE ABIOTIC ENVIRONMENT

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The fundamental niche delineates the environmental conditions where a given species/genotype can thrive in absence of biotic interactions (competitor, predators). One central tenet of evolutionary ecology is that the abiotic niche of a genotype results from its adaptation to these conditions. However, symbiosis raises the alternative possibility that a host's abiotic niche is instead determined by its interactor's adaptation to the environment. We investigate this question with gut microbiota as the symbiotic interactor. Specifically, our experiments on Artemia franciscana, (a small branchiopod from hypersaline environments), reveal that reduced survival at low salinity is caused by poor growth of gut microbiota. This effect is only seen when A. franciscana are fed with algae: axenic A. franciscana fed on yeast do not exhibit a drop in survival at low salinity. We provide further evidence that the gut flora is involved in the digestion of algae (the main food source of Artemia), and that its population dynamics are strongly affected by salinity. These findings shed a new light on the 'low salinity paradox' in this genus: because Artemia have a non-salty hemolymph (they are derived from freshwater branchiopods), they would be expected to perform well at low salinity. We propose that their performance at low salinity is in fact not determined by salt-related physiology, but by microbiota-mediated digestion of algae.

BACTERIAL COMMUNITIES INFLUENCED BY WOLBACHIA? BACTERIAL COMMUNITY STRUCTURE AND MAJOR PLAYERS IN THE TERRESTRIAL ISOPOD MICROBIOME

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Endosymbiotic communities, interactions between community members and their synergistic effects on host evolution have become important aspects in symbiosis research. Terrestrial isopods represent an excellent model system for multipartite symbioses due to their well-characterised association with Wolbachia. These endosymbiotic bacteria act as reproductive parasites in terrestrial isopods, inducing the feminisation of genetic males in Armadillidium vulgare. To date, three different feminising Wolbachia strains have been identified in this host and these strains vary in their distribution across host tissues. However, the microbiome of terrestrial isopods has never been analysed on a large scale and the role of Wolbachia within the bacterial community remains unknown. Here, we characterized the tissue-specific bacterial communities in A. vulgare, combining qPCR and 16S rRNA gene amplicon sequencing. To gain insight into intra-community dynamics, we analysed bacterial communities present in hosts from both laboratory and field populations, depending on (i) presence or absence of Wolbachia and (ii) infection with different Wolbachia strains. Wolbachia infection was an important factor influencing bacterial community structure. Furthermore, Wolbachia represented the predominant member of the bacterial community in infected individuals. These findings indicate that Wolbachia plays an important role within the terrestrial isopod microbiome. Apart from Wolbachia, we detected a second highly abundant bacterium: Candidatus Hepatoplasma crinochetorum, a facultative symbiont previously reported from the midgut caeca, was for the first time observed in all tested host tissues. Moreover, Ca. H. crinochetorum co-occurred with Wolbachia, although it was not observed in coexistence with all three Wolbachia strains. A better knowledge of the potential interactions of Wolbachia and Ca. H. crinochetorum will contribute to a better understanding of multipartite symbiotic interactions.

EVOLVING AS ALLIES? ECOLOGY AND EVOLUTION OF THE SYMBIOSIS BETWEEN BURKHOLDERIA BACTERIA AND TENEBRIONID BEETLES (LAGRIA SPP.)

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As suggested by the recently coined term "holobiont", the evolutionary history of an organism is determined by the selective pressures acting on a host and its symbionts as a whole, making it vital to consider both parts when addressing specific ecological and evolutionary questions. Among insects, the most diverse of animal groups, a considerable number of species live in close association with endosymbiotic bacteria. The association between beetles of the genus Lagria and bacteria identified as Burkholderia gladioli exhibits unique characteristics in the location and development of the symbiontbearing organs in the host. Although found in the microbiota of other insects, this is the first reported case in which Burkholderia, a genus with extraordinary metabolic and ecologic versatility, are harbored in specialized structures not associated to the insect gut. In female beetles of three Lagria species, we have identified closely related Burkholderia symbionts. Molecular analyses on one of these species (Lagria hirta) confirmed the presence of the bacteria in the larva and on the egg surface. Future results from egg-surface sterilization experiments, which allow the generation of aposymbiotic beetles, will reveal valuable information on host fitness effects related to symbiont presence. To obtain further insights on putative functions of the endosymbionts, we established in vitro cultures of the beetleassociated B. gladioli and subjected them to phenotype microarrays and genome sequencing. The results suggest potential participation in nitrogen recycling, aromatic compound metabolism and other detoxification functions. Ongoing comparative analysis of these data with closely related free-living and pathogenic Burkholderia will reveal genome modifications that could result from the endosymbiotic lifestyle, as well as features that might be relevant for the evolution and maintenance of this association.

INTRA-HOST PARASITE INTERACTIONS AND HOW THEY DEPEND ON THE PRESENCE OF A HOST

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Hosts and parasites are expected to influence each others evolution due to antagonistic interactions, potentially leading to host-parasite coevolution. However, many studies focus on the interactions between hosts and parasites ignoring that that within one host, different parasite genotypes may interact and may thus feed-back on the coevolution between host and parasite. The interaction between parasite genotypes may range from competition between genotypes for limited host resources to cooperation for more efficient host exploitation. Using *Caenorhabditis elegans* as host and the bacterial microparasite Bacillus thuringiensis we found indications for diverse interaction strategies between the bacteria. While they produce the toxin as a public good, they also show resource competition and spiteful behaviour by the production of bacteriocins. However, it remains unclear how stable these strategies are over the course of time, i.e. when hosts have to be repeatedly infected and when hosts may also adapt to these parasite strategies. For this reason, we performed a laboratory-based selection experiment in which a mixture of *B. thuringiensis* genotypes either coevolved with each other and the host or they just coevolved with each other without a host being present. After 48 generations parasites that coevolved with hosts were found to have a higher pathogenicity, and to be genetically more diverse, indicating that diversity is advantageous when coevolving with a host. Parasites that evolved without hosts were more likely to inhibit the growth of other *B. thuringiensis* clones while at the same time being more resistant towards inhibition by other clones, indicating a stronger antagonistic interaction between bacteria when they are not coevolving with hosts but just interacting with each other. Taken together these data provide novel insights into the interactions between parasite genotypes, how they evolve and how they depend on the presence of a coevolving host.

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IDENTIFYING SYMBIOTIC GUT-BACTERIA OF FUNGUS GROWING ANTS AND MAPPING THEM ON THE PHYLOGENETIC TREE

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Attine ants are good models for investigating complex biological interactions because every ant family is part of a symbiotic network with at least seven obligate participants: the fungus garden which is their primary food, Escovopsis fungus garden pathogens, cuticular Actinobacteria that produce antibiotics and their black yeast competitors, and two clades of nitrogen-fixing bacteria in gardens. However, there may be many more participants in the gut microbiome, but this community has essentially remained unexplored in spite of further mutualists being likely to occur there due to the highly specialized diets of the ants. We used 16S 454 pyrosequencing to identify gut bacteria in eight Panamanian genera of fungus-growing ants from both field and lab colonies. We complemented these data with FISH microscopy providing insight in the bacterial localization and possible roles of some of these bacteria in the ant-fungus mutualism. Mapping some of the prevalent OTUs on the attine phylogenetic tree showed a number of distinct patterns that appear consistent with major transitions in social and symbiont evolution.

THE INFLUENCE OF NATURALLY ASSOCIATED MICROBES ON CAENORHABDITIS ELEGANS LIFE HISTORY

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More or less all organisms, ranging from sponges to humans, associate with an often extremely diverse microflora. Microbial associations are known to be of immense importance for a host's development, immunity, and life history - all biological fields where the bacteriovorus nematode Caenorhabditis elegans has been studied with great success. On the other hand, laboratory cultures of the worm are axenic or monoxenic by default and we lack basic knowledge about its natural ecology; we even do not know what the nematode feeds on in nature. As a result, complex microbial interactions have mostly been created artificially by studying human pathogens or simply ignored. To rectify this discrepancy, we here used 16S rDNA deep sequencing of natural samples in combination with 16S rDNA Sanger sequencing of culturable bacteria to demonstrate a rich and diverse microflora associated with *C*. elegans. The prominent identified orders included Bacteroidetes (Flavobacteriales, Sphingobacteriales), Proteobacteria (Rhizobiales, Pseudomonadales, Enterobacteriales) and Firmicutes (Lactobacillales). We furthermore characterized the exact relationship between individual bacterial strains and the nematode host, using fitness assays, behavioral tests, differential interference contrast microscopy, and fluorescence *in situ* hybridization. Our analysis revealed high nematode fitness on Gammaproteobacteria, whereas Bacteroidetes and Proteobacteria were over time generally more attractive than Actinobacteria and Firmicutes. Our project combines the power of *C. elegans* as a model organism with its natural ecology to establish a tractable genetic model system for the in-depth analysis of naturally occurring host-microbiota interactions.

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DYNAMICS OF PROTECTIVE SYMBIOSIS

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Symbiosis, in which different species engage in prolonged and intimate associations, is gaining recognition as a ubiquitous feature of animal life. In many species, associations with symbiotic microorganisms are pervasive. These microbial associates are often heritable, transmitted with high fidelity from parent to offspring. Because host species and their symbionts share fates, inherited symbionts may exert beneficial effects on the hosts like conferring protection against adverse conditions. Among insects, aphids represent the best-studied case of protective symbioses. These sapfeeding insects may harbour one or several heritable bacterial symbionts, some of them providing protection against various natural enemies. To understand the dynamics of these protective symbioses in host populations, two barriers have to be investigated: the ability of microbial symbionts to infect a new host individual and the maintenance of symbionts infection over host generations. Aphids' biotic environment includes both the plants they feed on and the natural enemies they encounter (predators and parasitoids). Aphid individuals harbouring or not protective symbionts may co-occur on common plants, and horizontal transfers (transmission between host individuals) could potentially occur through direct contacts between aphids and/or the shared plant. Once present in a host population, the maintenance of protective symbionts depends on costs and benefits associated with such a symbiosis. In nature, selection pressures exerted by natural enemies may strongly fluctuate. By affecting host's ecology through protective phenotype, the evolutionary relationship between a host and its protective symbionts would therefore be temporally and spatially dynamic. Horizontal transmission of protective symbionts and their maintenance in host populations, studied by various empirical approaches, will be presented to contribute to our understanding of symbionts' dynamics in natural host populations.

COMPARATIVE GENOMICS OF INSECT ENDOSYMBIONTS EXTRACTED FROM SINGLE HOSTS

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Insect endosymbionts represent the paradigm of bacterial adaptation to a host-dependent life and their study contributes to our understanding on microbial ecology and genome evolution. Their informative nature is nonetheless counterpoised to their inaccessibility. Here we demonstrate that single-host endosymbiotic communities can be studied from next-generation sequencing data after purification and whole-genome amplification of endosymbionts. We focus on the comparative genomic study of two samples belonging to pure *Hamiltonella defensa* sequences extracted from two individual aphids from the tribe Macrosiphini. The assembly of their genomes show that they are 1.8 and 2.0 Mb-long, respectively, and their gene content is shown to be similar to that of other *H. defensa* genomes. We used this new data to fully resolve the phylogenetic relationships between the known *H. defensa* and related enterobacteria, and to reconstruct the history of gene gains and losses in their evolutionary lineage. Finally, we performed an in-depth analysis of the genome content and evolution of the APSE bacteriophage, inserted in the *H. defensa* genomes, which has a remarkable ecological importance in protecting the aphid host against parasitoid wasp infections.

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FLORAL DISPLAY DIFFERENTIALLY AFFECTS SIRING SUCCESS AND SEED PRODUCTION IN THE INSECT-POLLINATED HERB PRIMULA FARINOSA

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A prominent floral display may be favoured by selection because it increases attractiveness to pollinators, but may be associated with a cost in terms of an increased risk of damage from seed predators and grazers. While the benefits of a large display may be expressed through both male and female function, the negative effects of herbivore attack may often appear only after pollen dispersal, and thus predominantly influence female reproductive success. The short-lived, hermaphroditic herb *Primula farinosa* is dimorphic for scape length and occurs as a long-scaped and a short-scaped morph. The long-scaped morph displays its flowers well above the ground, whereas the short-scaped morph displays its flowers close to the ground. We conducted a field experiment with genotyped plants at two sites in SE Sweden to examine whether male reproductive success of the long-scaped morph is greater than that of the short-scaped morph, and whether this may compensate for a higher risk of reduced seed production caused by interactions with seed predators and grazers. Female reproductive success was assessed by quantifying seed production and male siring success by genotyping offspring. At the first site, where grazing pressure and seed predation were high, the long-scaped morph sired significantly more offspring but produced fewer seeds than did the short-scaped morph. At the second site, the two morphs did not differ significantly in siring success or seed production. The results demonstrate that interactions with mutualists and antagonists can differentially affect selection on floral display through male and female function and produce a relationship between floral display and plant functional gender. Positive effects of a large floral display on male reproductive success may at least partly compensate for an increased risk of herbivore-mediated reduction in seed production.

D23SY04PS0758

METAGENOMIC ANALYSIS OF GUT BACTERIA FROM THE RED FLOUR BEETLE TRIBOLIUM CASTANEUM

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The relevance of symbiotic microbial communities in the gut is increasingly being studied in a large number of animal species, from sponges to primates. Although the red flour beetle *Tribolium castaneum* represents a well-established experimental model organism for studying ecological, evolutionary and developmental topics, to the best of our knowledge, there are no studies on the composition of the gut microbiota of this insect. We examined the bacterial composition of the digestive tract of *T. castaneum*. For this, two approaches were combined: a culture-independent metagenomic analysis based on 16S rRNA sequences and a classical bacterial cultivation method. The comparison of bacterial 16S rRNA sequences between guts of larval and adult beetles revealed a generally lower diversity of bacterial genera, compared to other insect species. Moreover, the diversity of bacterial genera was higher in guts of adults than in larvae. As expected, cultivation of gut contents on different growth media confirmed only a minor part of the genera found in the metagenomic analysis. The information on bacterial communities in the gut of *T. castaneum* will be useful for future studies testing interactions between *T. castaneum* and its microbiota, potential symbionts and pathogens.

D23SY04PS0893

CULTURE-INDEPENDENT CHARACTERISATION OF THE CORE GUT MICROBIOME OF FUNGUS-GROWING TERMITES

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Fungus-growing termites (subfamily Macrotermitinae, family Termitidae) live in an obligate mutualistic symbiosis with the fungus Termitomyces. All other termites rely on gut microbes for the breakdown of plant material and other forage, and it has been generally assumed that the association with Termitomyces has reduced the need for fermentative gut microbes after the Macrotermitinae became fungus-farmers. Only few studies have explored this in any detail and the identities, levels of interaction-specificity with the termite host, and consistency in bacterial communities between host species have remained largely unknown. Here, we employ bacterial 16S rRNA 454 high-throughput pyrosequencing to identify a potential core microbiome in the fungus-growing termites - i.e., a distinct set of bacteria present across lineages in the termite phylogeny. Comparative analysis of 9 fungusgrowing termite species from 5 genera suggests that a core gut microbiome indeed exists, as all bacterial taxa of high abundance were present in all termite species examined. However, quantitative differences in microbiome composition between termite species and genera were also noticed, possibly associated with differences in substrate use and *Termitomyces* lineage reared. Our results are consistent with major changes in gut microbiomes having occurred when fungus farming evolved 30 MYA, followed by relatively modest elaborations in response to ecological conditions. This might help explain why neither the termites nor Termitomyces ever abandoned the symbiosis or teamed-up with another termite of fungal partner lineage.

CHRYSOMELINA LEAF BEETLES PROFIT FROM WIDELY EXPLOITING THEIR HOST PLANTS: ABC TRANSPORTERS ARE INVOLVED IN THE SEQUESTRATION OF POISONOUS PHYTOCHEMICALS

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The ubiquitous consumption of plants by insect herbivores requires in many cases detoxification of noxious phytochemicals. One of the most ingenious detoxification strategies widespread in insects is sequestration which involves uptake, transfer and concentration into specialized tissues. Frequently, sequestered compounds are not only stored securely by the insects, but further; insects evolved the ability to use an overwhelming diversity of exogenous compounds for their own benefit within the competitive interactions in natural ecosystems, like for repelling insects' enemies. Here, we report the first example of a transport protein crucial for the translocation of phytochemicals in insects. The transporter, referred to here as *Cp*MRP, belongs to the ATP binding cassette (ABC) transporter family and functions in the defensive exocrine glands of juvenile poplar leaf beetle, Chrysomela populi, for shuttling plant derived metabolites into deterrent secretions. Silencing of CpMRP in vivo by using RNAi creates a defenseless phenotype which indicates its key function also for the secretion process. Our analyses of the transport activity suggest that involving a broad-spectrum carrier in the sequestration of phytochemicals may be advantageous for insects to affiliate alternative host plants. Indeed, we identified transporter sequences highly similar to *Cp*MRP in the larval glands of a related Chrysomelina leaf beetle species which forms allopatric populations and shifts hosts between chemically different plant families. This demonstrates the importance of the substrate flexibility provided by ABC transporters to avoid feeding specialization and host-derived chemical defense leading to an evolutionary dead end. Moreover, ancestral species of *C. populi* synthesizing defensive compounds endogenously already possess in their glands CpMRP-similar proteins which shed new light on the evolution of sequestration in the taxon Chrysomelina.

FRIENDS OR FOES: ECOLOGICAL RELATIONSHIP DICTATES SPATIAL STRUCTURING IN POLYMICROBIAL COMMUNITIES

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Polymicrobial communities usually form surface-attached and spatially structured consortia such as biofilms. These communities display a broad range of metabolic interactions, thereby setting the stage for the emergence of diverse ecological outcomes, spanning competition, exploitation, or mutualism. Our understanding, however, of how the mechanistic nature of interspecific interaction shapes spatial structure within these communities is still limited. Using an individual-based model of a two-species community growing on a surface and where resources are traded for detoxification, we explore the relationship between mechanism of interspecific interaction and emergence of spatial structure within the community. We show that both abiotic and biotic factors can affect the spatial organization of species within these polymicrobial communities, and in a manner that reflects the balance between the costs (interspecific competition) and benefits (need for help) of association. Understanding the mechanisms that shape the emergence of spatial structuring within multi-species communities may provide new insights into how to maintain beneficial polymicrobial communities (e.g. microbiota) and combat polymicrobial infections.

THE EVOLUTION OF CUTICULAR HYDROCARBON PROFILES IN ANT-ANT ASSOCIATIONS

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The insect cuticle is covered with hydrocarbons, which form complex profiles of up to 100 different substances. These hydrocarbons protect against desiccation, but also serve as recognition cues. Especially in social insects, cuticular hydrocarbon profiles carry information on species and colony membership, sex, and caste. They vary greatly between insect species, but the evolutionary cause for this diversity is largely unknown.
In social insects, nestmates are recognized by their cuticular profile, and individuals with deviant profiles are attacked. Thus, ant species that are commensals, parasites or mutualists of other ant species need to adapt their chemical profiles to be tolerated. We investigated the influence of interspecific associations on chemical profiles in the ant genera Camponotus and Crematogaster. Associations between species of these two genera occur world-wide and vary in intimacy and interspecific aggression. Aggression between species varied with association type, and corresponded to different traits of the hydrocarbon profiles. In associations in which two species share a nest (parabioses), both species showed little interspecific aggression, and possessed long-chained hydrocarbons, coupled with a shift to methylbranched alkenes and alkadienes. In a Crematogaster-Odontomachus parabiosis, interspecific aggression was higher and the involved Crematogaster species exhibited shorter hydrocarbons. Camponotus species in trail-sharing associations possessed unusually simple profiles, or ones with high chain lengths or high amounts of nalkanes. We also found evidence for physiological and climatic constraints on hydrocarbon profiles. Notably, chemical diversity in parabiotic species was extremely high, suggesting a rapid evolution of profiles, and possibly triggering speciation. Overall, associated ant species exhibit cuticular hydrocarbon profiles that are shaped by the need to be tolerated, but they employ different chemical tolerance strategies.

MACROEVOLUTION OF HELICONIINI BUTTERFLIES IS STRONGLY CORRELATED WITH DIVERSIFICATION OF PASSIFLORACEAE HOST PLANTS

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The famously variable wing patterns of neotropical *Heliconius* butterflies (Nymphalidae: Heliconiini) advertise toxicity acquired by obligatory feeding on cyanide-rich Passifloraceae vines. Heliconius and the other nine genera of Heliconiini constitute the main group of Passiloraceae herbivores. Although studies of individual associations abound, a systematic analysis of this coevolutionary relationship has never been attempted. I present a comprehensive Bayesian phylogeny of New and Old World Passifloraceae based on publicly available data from 13 plastidial, mitochondrial and nuclear markers, comprising 15.5 kbp of sequence. The tree is compared to a previously estimated multilocus phylogeny of Heliconiini and both are dated using relaxed molecular clocks. The final dataset captures 65% of all the known Heliconiini- Passifloraceae relations. A distance analysis (ParaFit) demonstrates a strong signal of coevolution and shows that 45% of all the associations are due to evolutionarily conserved feeding preference and not recent host switches. Cost model simulations in Jane 4 also support the hypothesis of coevolution, suggesting frequent butterfly speciation events with and without subsequent host switches. Contrary to traditional views, I show that the number of host plants used by a heliconian depends strongly on the range of the species, but not on its systematic position. However, the extent of evolutionary conservatism in host plant choice varies between the major butterfly clades: Heliconius stand out from other genera as highly co-evolved. My study provides strong evidence for the diversification of Passifloraceae as a significant factor in the macroevolution of Heliconius, an emerging model group for evolutionary genomics. It also provides testable hypotheses for the ongoing studies of butterfly molecular adaptation to the host plants at the genome level.

INTENSE COMPETITION BETWEEN ARBUSCULAR MYCORRHIZAL MUTUALISTS IN AN IN VITRO ROOT MICROBIOME NEGATIVELY AFFECTS TOTAL FUNGAL ABUNDANCE

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The root microbiome is composed of an incredibly diverse microbial community that provides services to the plant, including nutrient provision, increased stress resistance and pathogen exclusion. A major question in rhizosphere research is how species in root microbiome communities interact with each other, and their host. In the nutrient mutualism between host plants and arbuscular mycorhizal fungi (AMF), hosts exchange carbohydrates for mineral nutrients from the fungal partner. While there is high variation in the benefits AMF species provide to their host plants, we still know very little about how this variation drives fungal community composition and competition within the root microbiome. Here we use an artificial root system (in vitro root organ cultures) to investigate competitive interactions between a previously characterized 'high' and 'low' quality AMF species, under standard and high phosphorus availability. We show that negative competitive interactions between AMF species reduces overall fungal abundance by nearly 5-fold in the host root, and that negative interactions are less strong in the extraradical mycelial network. We find that the high-quality species, Rhizophagus irregularis, consistently outcompete the lower quality species, Glomus aggregatum, both inside and outside the host root. However *G. aggregatum* is not completely eradicated and still is able to reduce the abundance of *R. irregularis*. Phosphorus availability did not change the outcome of these interactions. We suggest that in vitro root systems offer a novel platform to study ecological dynamics in the root microbiome under precisely controlled conditions.

BARGAINING POWER AND A BIOLOGICAL MARKET ANALYSIS OF THE PLANT-MYCORRHIZAL SYMBIOSIS

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Mutualistic cooperation can easily be stabilized when one partner is in relative control, interacts with a number of potential partners from the other species, and has some mechanism to enforce cooperation. In contrast, in the mycorrhizal mutualism, there is no apparent partner 'in control'. Here, both partners may interact with multiple symbionts simultaneously. This complex series of interactions means that neither partner determines the terms of the interaction. Biological market theory models offer an approach for modeling and conceptualising such coevolutionary interactions between multiple partners. However, previous biological market models have made two assumptions that limit the extent to which they can be applied to mutualisms such as that between plants and fungi. First, they assumed that markets consist of an infinite number of partners. This prevents any partner having the bargaining power that comes from being one of a limited number of partners. Second, they assumed that cooperation is stable, rather than asking when it would be stable. Consequently, we have developed and analysed a market model without these assumptions and thus more readily applicable to the mycorrhizal mutualism. First, we found that whether or not individuals engage in mutualistic trade depends both qualitatively and quantitatively upon the number of partners of each species, as well as their degree of specialisation. Second, we found that, in a market model, mutualistic trade implies that at least one species relies completely on the other for at least one resource. Finally, we found that incorporating bargaining power may mean that many of the potential gains from trade are not exploited.

COMPARATIVE ANALYSIS OF SYMBIOTIC COMMUNITIES IN HIPPOBOSCIDAE

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Symbioses between bacteria and insects can range from simple bilateral relationships, with a single obligate mutualist, to multilateral models with complex bacterial communities. In hematophagous hosts, typical examples of such associations are provided by the symbiosis of human body louse Pediculus humanus with its primary symbiont Rieisa pediculicola, and tsetse fly Glossina morsitans with bacterial community containing a primary symbiont *Wigglesworthia glossinidia*, a facultative associate Sodalis glossinidius and Wolbachia. To allow for broader comparison of the symbiotic communities and their effect on the biology of different hosts, we analyzed symbiotic bacteria in hosts closely related to Glossinidae, the louse flies of the family Hippoboscidae. These organisms share many unique biological characteristics with Glossinidae, e.g. strict hematophagy, adenotrophic vivipary and nourishment through milk glands, and a specialized midgut section (bacteriome). Using Illumina sequencing, we characterized genomes of the symbionts associated with two biologically different species, namely *Melophagus ovinus* and *Lipoptena fortisetosa*. We demonstrated that the complexity of the associations differs among these species. In analogy to tsetse symbiosis, *Melophagus ovinus* carries complex symbiotic community involving obligate mutualist from the genus Arsenophonus (Gammaproteobacteria), a facultative symbiotic bacterium originated within Sodalis lineage (Gammaproteobacteria), a widespread bacterial associate of the genus *Wolbachia*, and two additional microorganisms Bartonella melophagi (Alphaproteobacteria) and Trypanosoma melophagium (Excavata). In contrary, *Lipoptena* represents a model of bilateral symbiosis harboring *Arsenophonus* bacteria as a sole symbiont. We discuss possible consequences of the symbiotic community structure on the metabolic functions within the host.

D23SY04PS1299

THE BACTERIAL COMMUNITY OF SOCIAL SPIDERS

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The spider genus Stegodyphus is characterized by three independent origins of sociality where spiders form colonies by constructing a communal web. Mating occurs within the colony resulting in high levels of inbreeding and consequently in low genetic variation between colony members, moreover, colonies exhibit a highly female biased sex ratio. The persistence of these characteristics is challenging to explain from an evolutionary point of view, not in the least as the extent to which these traits are influenced by external factors remains unknown. Especially the bacterial community can potentially have profound effects on the evolutionary dynamics of sociality. Pathogenic bacteria can infect and spread rapidly in a colony due to aggregated group living and low genetic variation, explaining the observed, strong metapopulation dynamics in social species. The female biased sex ratio could in part be explained by the effects on reproduction by maternally inherited endosymbiont bacteria such as Wolbachia, Rickettisa,... As a first approach to elucidate this interaction we characterized the bacterial community using next generation sequencing techniques in social and solitary sister species. This provides us with a detailed overview of the bacterial diversity related to different levels of sociality and allows to identify key bacterial species that play a role in the evolution of sociality in spiders.

MICROBIOTA, VERTICALLY TRANSMITTED INFECTIONS, AND DIAPAUSE IN DAPHNIA

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The roles of communities of host-associated microorganisms (microbiota) in the function, ecology and evolution of their hosts are increasingly being demonstrated across different model systems. We have recently shown that in the water flea *Daphnia*, a well-studied model for many questions in evolutionary ecology, absence of microbiota significantly decreases survival, growth and fecundity compared to animals with normal microbiota. In this project, we examine the influence of the microbiota on the severity of an infection caused by a microsporidian that is vertically transmitted into diapausing eggs of *Daphnia* by raising infected and uninfected *Daphnia* with and without microbiota. Since previous observations suggested that daphnids emerging from diapausing eggs obtain their microbiota horizontally from the environment, we also specifically investigated the ability of several environmental bacteria to restore the normal functioning of infected and uninfected animals. This project addresses both the interaction of parasites and mutualists during the course of the organism's lifetime, and the particular challenges of recruiting and maintaining a functional microbiome faced by diapausing organisms.

D23SY04PS1389

SPECIFICITY AND TRANSMISSION OF BACTERIAL ENDOSYMBIONTS IN MEGALOMYRMEX SOCIAL PARASITES AND THEIR FUNGUS-GROWING ANT HOSTS

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Communities or single lineages of bacterial symbionts are increasingly recognized as major fitness determinants of insects, and they are particularly likely to be mutualists when host diets are specialized and partially deficient. Some insect hosts have independently domesticated related microbes to meet similar challenges, but whether hosts with different life histories can maintain the same symbionts merely by sharing the same food niche has not been explicitly investigated. The nest-sharing symbiosis between Megalomyrmex social parasites (Formicidae: Solenopsidini) and their fungus-growing ant hosts (Formicidae: Attini) provides a unique opportunity to address this question, as both lineages rely on the same narrow fungal diet, but are only distantly related. We used a combination of 454 pyrosequencing and diagnostic PCR screening to map the diversity of bacteria associated with Megalomyrmex ants across their phylogenetic tree, which also contains clades of free-living generalist predators. We show that a specific group of Entomoplasmatales is associated with socially parasitic 'thief-ant', 'agropredator' and 'guest-ant' lineages of *Megalomyrmex* and appears to be derived from and always shared with the attine host ants. Moreover, the microbiotas of guest-ants appear to be completely dominated by a specific Bartonellaceae lineage, which can but does not need to be accompanied by the Entomoplasmatales bacteria. Bartonellaceae are native to the guest-ants and are secondarily transmitted to some (ca 40%) but not all host colonies. Our results suggest that host and socially parasitic ants contribute their own vertically transmitted bacteria to their permanent symbiosis and that these bacteria can be asymmetrically transmitted to the partner species via the fungus-garden that they share. The function of these endosymbionts remains unknown.

MICROBIOTA OF AVIAN BROOD PARASITES SHAPED BY FOSTER PARENT SPECIES: A ROLE IN ENHANCED IMMUNITY OF COWBIRDS?

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Avian brood parasites that exploit many species (extreme host-generalists) have more effective immune responses than related, non-parasitic species. This innovative trait, enhanced immunity, has supported invasion of new habitats and exploitation of new host species with the result that the North American brown-headed cowbird has accomplished a continental-scale range expansion well beyond its ancestral range in the central American prairies. Given the symbiotic role of the gut microbiome in vertebrate immunity, we used next generation sequencing to examine gut and cloacal microbe communities of the cowbird and assess whether microbial inoculations from foster parent species may contribute to greater diversity of cowbird microbiomes. We found that indeed cowbird cloacal beta diversity (among individuals) is higher than beta diversity of a closely-related, non-parasitic blackbird species and that the single most significant predictor of cloacal diversity is gut diversity of the same individual. Our results are consistent with the idea that biological interactions between cowbirds and their facultative parent species contributed to higher diversity in cowbird microbiota at the cowbird community level, and this in turn has contributed to the evolution of cowbirds' more robust immunity

Symposium

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

20 and 21 August



Program

Tuesday 20 August

Session(s): 1, 2, 3, 4 Wednesday 21 August Session(s): 5, 6, 7, 8

Organisers: Patricia Gibert and Patrícia Beldade

Invited speakers: Jacintha Ellers and Anthony Zera

Description:

Phenotypic plasticity refers to the ability of a genotype to produce different phenotypes in different environments. Its study integrates disciplines analyses multiple and at all levels of biological organization; from the molecular regulation of changes in organismal development, to variation in phenotypes and fitness in natural populations. The symposium welcomes work searching to understand how changes in external environment affect (or not) phenotype, how alternative phenotype perform in distinct environmental conditions, and how that weighs into account for the evolution of plasticity.

D20SY05IT10:30R1

A NEW PERSPECTIVE ON PHENOTYPIC PLASTICITY: TAKING INTO ACCOUNT PHYSIOLOGICAL MECHANISMS CHALLENGES CLASSIC PLASTICITY THEORY

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Phenotypic plasticity is ubiquitous but we have poor knowledge about the underlying mechanisms. The reason for this is that classical studies of phenotypic plasticity developed and tested theory only for organismal traits, such as morphology or life history. However, a new and upcoming perspective on plasticity encompasses also transcriptional and physiological flexibility in an effort to study the underlying mechanisms of phenotypic plasticity. This raises the question how plasticity at different organisational levels interacts to produce the optimal phenotype in different environments. Here, I will focus on the evolution of temperature-induced plasticity as a case study to show that greater phenotypic plasticity at one organisational level is associated with environmental canalization (lack of plasticity) at the other level. More specifically, my work shows that strong physiological flexibility in response to temperature correlates with low sensitivity to temperature for fitness traits. In this context, I will discuss costs of plasticity and the evolution of plasticity as a means of adaptation to changing thermal conditions. I will also identify candidate physiological pathways underlying variation in thermal response. Ultimately, taking into account the mechanism underlying plasticity will challenge the classical dichotomy between phenotypic plasticity and environmental canalization. Instead, the key question is at what level of biological organization phenotypic plasticity will evolve.

D21SY05IT10:30R1

MORPH-SPECIFIC CIRCADIAN PLASTICITY AND G X E FOR JUVENILE HORMONE AND GENE EXPRESSION IN A WING-POLYMORPHIC CRICKET

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The endocrine basis of phenotypic plasticity in insects has been the subject of considerable study and speculation for decades. Yet, detailed information on endocrine mechanisms is still uncommon, and many "classic" examples of endocrine plasticity are, in fact, only weakly supported by experimental data. Furthermore, an important aspect of plasticity, circadian plasticity, has remained virtually unstudied. During the past decade, the Zera laboratory has undertaken detailed studies of morphspecific JH titer variation in the wing-polymorphic cricket, Gryllus firmus. This species exhibits a classic life history trade-off consisting of a dispersing morph (LW; long-winged) that delays reproduction and a flightless morph (SW; short-winged) that exhibits enhanced early-age fecundity. An unanticipated and dramatic genotype (morph) x environment (time-of-day) interaction for the blood JH titer (i.e. genetic variation for circadian plasticity) was identified in this species. In the LW morph the JH titer rises and falls 20-100 fold within a few hours before and after lights-off in the laboratory or sunset in the field. Failure to take this rhythm into account can result in substantial errors in interpreting the role of JH in morph adaptation. The morph-specific JH titer rhythm appears to be common in cricket species; the JH cycle likely regulates some aspect of flight, which only occurs in the dispersing morph at night. A recent transcriptome study identified a morph-specific circadian rhythm with respect to global gene expression. Daily change in transcript abundance occurs for a greater number of genes in the LW than in the SW morph, and is more prominent than differences in expression between LW and SW morphs at the same time of day. Circadian plasticity and G X E are potentially common, important, but largely unstudied aspects of phenotypic plasticity and life history adaptation. Supported by NSF award IOS – 1122075 to AJZ

D20SY05RT11:18R1

CAN WE PREDICT THE EFFECT OF THERMAL FLUCTUATIONS ON SPECIALIST AND GENERALIST REACTION NORMS?

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Reaction norms depict the environmental effects on phenotypic traits and are used to predict the global change consequences on organisms. However, studies performed at constant temperatures have limited ecological significance because expressed phenotypes depend on the range and frequency of environmental states. By using the Jensen's Inequality (a mathematical property of nonlinear functions), we predict that the effect of thermal fluctuations on the phenotype depends on the shape of the reaction norm: a negative effect of the thermal fluctuations when the reaction norm is convex and a positive effect when the reaction norm is concave. This study measures the impact of diel fluctuations in developmental temperature on phenotypic expression of traits related to fitness and energetic resources in two strains of the parasitoid wasp Venturia canescens differing in their thermal sensitivity. In a first experiment, we compare the effect of a constant versus a fluctuating thermal regime having the same means (20, 25 and 30 °C) on reaction norms of life history traits and of energetic reserves. In a second experiment, we examine the effects of a natural thermoperiod in field on these traits. As predicted, our results show that the shape of the reaction norm defines the phenotypic changes induced by the development under fluctuating thermal conditions. Moreover, our results emphasize the significance of taking into account several phenotypic life history traits to study the adaptive value of phenotypic plasticity. We also show that the level of energetic resources depends on the mean developmental temperature and not on the thermal regime. Finally, the field experiment confirms that the phenotype of these parasitoids depends on the temperature variation. This is the first experimental study demonstrating that Jensen's Inequality can quantitatively predict the effect of thermal fluctuations on life-history traits of an ectotherm species.

D20SY05RT11:42R1

WHEN IN DOUBT, HEDGE YOUR BETS. DIVERSIFICATION AND PHENOTYPIC PLASTICITY IN SPIRODELA POLYRHIZA (L.) (GREATER DUCKWEED)

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Reliable environmental cues may lead to the evolution of adaptive phenotypic plasticity. However, when cues are unreliable and the environment is thus unpredictable, so-called "bet hedging" is expected to evolve. Bet hedging characters decrease variance in fitness, and short-term success (i.e. expected fitness) is sacrificed for success over generations. Empirical support for bet hedging is limited, but this mode of response is increasingly being recognized as an important response to environmental variance. Here, the phenology of turion formation in greater duckweed (*Spirodela polyrhiza* (L.)) is used to study bet hedging and phenotypic plasticity in the field. Turions are heavy, non-vegetative fronds that sink into the sediment, allowing this free-floating hydrophyte to persist through unfavourable conditions. Mean days to turion formation was found to differ among pond environments-fronds from ponds of greater unpredictability had greater variability in the timing of turion formation. In a field transplant study, fronds from the more predictable pond environmental variability. The success of *S. polyrhiza* may be attributed to evolved strategies, which are dependent on the availability of dependable cues. This study provides evidence of the occurrence of both bet hedging and plasticity in fluctuating pond environments.

D20SY05RT14:00R1

PHENOTYPIC DIVERGENCE AMONG SPADEFOOT TOAD SPECIES REFLECTS ACCOMMODATION OF MECHANISMS UNDERLYING DEVELOPMENTAL PLASTICITY

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Selection in heterogeneous environments favours plasticity as it allows organisms to adapt to rapidly changing conditions. Developmental plasticity allows populations to withstand rapid environmental changes and confers an overall faster rate of adaptation. Conversely, if plasticity costs are high and the environment stabilises, selection results in genetic assimilation, which could result in trait divergence and species diversification. Current evolutionary theory contemplates that phenotypic divergence between species may initiate as environmentally-induced expression of alternative phenotypes. Descendant lineages of a plastic ancestor evolving in stable divergent environments may lose plasticity over time, their development becoming specialised to produce fixed phenotypes matching each environment. In that case, we would expect ancestral plasticity to mirror differences among taxa and that the same mechanism allowing ancestral plasticity was also the main mechanism explainig species divergences. In that light, we are studying mechanisms of plasticity behind the evolutionary divergence of spadefoot toads. Old World species (*Pelobates*) breed in long lasting ponds and have long but plastic larval periods, whereas New World species (Scaphiopus) have specialised in ephemeral ponds and have evolved very short larval periods. We hypothesise that *Scaphiopus* has undergone genetic accommodation of ancestral plasticity, which has resulted in canalised short larval periods. To test this hypothesis we have studied the mechanisms underlying developmental acceleration in response to pond drying and compared it across species. We have found that *Pelobates* tadpoles, which reflect the ancestral state of the group, increase their metabolic rate, and thyroid hormone and corticosterone concentrations in response to decreased water levels. All these parameters, however, seem to have been canalised in *Scaphiopus*, lending support to the hypothesis of genetic accommodation.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

D20SY05RT14:24R1

EXPERIMENTAL EVOLUTION OF PLASTICITY IN A VIRUS

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Coinfection of parasite genotypes can select for various changes in parasite life history strategies, with consequences for disease dynamics and severity. Here we show adaptive phenotypic plasticity evolving in real time in response to coinfection under conditions in which both single infections and coinfections are common. We experimentally evolved an obligate-killing virus under conditions of single virus infections (single lines) or a mix of single infections and coinfections (mixed lines) and found mixed lines to evolve a plastic lysis time: they killed host cells more rapidly when coinfecting than when infecting alone. This behaviour resulted in high fitness under both infection conditions. We also discuss how population structure and the importance of within-patch prudence affects the fitness and virulence of populations of these viruses. Such plasticity and prudence has important consequences for the epidemiology of infectious diseases and the evolution of cooperation.

D20SY05RT14:48R1

DIVISION OF LABOUR THROUGH PHENOTYPIC HETEROGENEITY IN CLONAL BACTERIAL POPULATIONS

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Individual bacteria in clonal populations often vary considerably in the expression of certain traits, even under uniform environmental conditions. At the proximate level, such phenotypic heterogeneity is generally attributed to stochastic processes that generate noise in gene expression. At the ultimate level, it remains often unclear whether phenotypic heterogeneity is beneficial and can be selected for. Here, we tested this possibility by studying between-individual variation in the production of a cooperative public good in the bacterium Pseudomonas aeruginosa. We show that phenotypic heterogeneity can provide benefits because it allows bacteria to realise an efficient form of division of labour. Specifically, we found that the production of public good followed an economy of scale: for the individual cell, the cost per public good molecule decreased with higher production rates. Accordingly, population-level costs of public good investment are minimized when only a fraction of the cells produces most of the public good, whilst benefits are equally shared among all individuals in the population. However, our data also indicate that the degree of division of labour is constrained by both the genetic architecture of the trait, and the overall need for the public good, which in turn depends on prevailing environmental conditions. Taken together, our data suggest a form of adaptive phenotypic plasticity, where individuals adjust their phenotype not only in response to environmental changes, but also in response to the population-level costs associated with the expression of a cooperative behaviour.

D20SY05RT15:12R1

FISH FARMS SELECT FOR INCREASED PHENOTYPIC PLASTICITY IN GROWTH AND VIRULENCE IN A FISH PATHOGEN

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Opportunistic pathogens generally face two vastly different environments - within the host and outside host. One mechanism allowing for adaptation to alternating environments is switching between phenotypes (phenotypic plasticity). The opportunistic fish pathogen *Flavobacterium columnare* can be found from natural waters and from fish farms and it exhibits two reversible colony morphologies; a non-virulent "rough" and a virulent "rhizoid" morphology. As compared to natural waters, fish farms can be considered as extreme environments in terms of available host resources, but also in terms of stress caused by chemical and antibiotic treatments. Fish farms could thus be expected to impose higher selection pressures for coping between the within and outside host environment, and to select for increased phenotypic plasticity. To test these ideas we measured growth parameters of rhizoid and rough colony morphotypes of *F. columnare* isolates both from natural waters and from disease outbreaks at fish farms in different resource concentrations and temperatures, and tested their virulence with a zebrafish challenge model. We found that the non-virulent "rough" morphotypes had a higher growth rate and lower virulence than the "rhizoid" morphotypes, but only if the isolate was originating from the fish farms. This suggests that phenotypic plasticity between two morphotypes of opportunistic pathogen and their characteristic traits is clearly selected for in fish farms rather than in the natural environment.

D20SY05RT15:45R1

WHEN TO SHRINK? UNDERSTANDING THE EVOLUTION OF NEGATIVE GROWTH IN ECTOTHERMS

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Body size is a key trait linked to the main determinants of fitness: fecundity and mortality (1). For many ectotherms, adult body size is plastic and follows the temperature size rule: individuals grow smaller at higher temperatures (2). Adapting the body size to the environmental condition is crucial and species with continuous growth such as fish and ametabolous hexapods benefit from their ability to adjust their size throughout life (3). But growing larger is in general considered to be the only option for theses organisms whereas shrinking could be advantageous especially if the organism has to cope with a temperature increase. We experimentally demonstrate that such shrinking occurs in the Collembola Folsomia candida - an ametabolous hexapode which continues moulting during its whole life - when they undergo a temperature increase. Comparing multiple clonal strains, we demonstrate genetic variability for this ability to shrink suggesting that negative growth can evolve under natural selection. Using a theoretical model fitted to the collembolans life cycle, we found that for predictable temperature variation, shrinking evolves for environmental temperature variations happening at a time scale close to the individual's lifetime. For stochastic temperature variation, negative growth can also be selected, as a way for large individuals to escape high mortality following a temperature increase and therefore avoid being stuck in a so called "size ratchet". 11. Kingsolver JG, Huey RB. Size, temperature, and fitness: Three rules. Evolutionary Ecology Research 2008;10(2):251-68. 2. Atkinson D, Sibly RM. Why are organisms usually bigger in colder environments? Making sense of a life history puzzle. Trends Ecol Evol 1997;12(6):235-9. 3. Charnov EL. Size and temperature in the evolution of fish life histories. Integr Comp Biol 2004, Dec;44(6):494-7.

D20SY05RT16:09R1

PLASTIC TRAITS OF WHITEFISH DEPENDENT ON THE INTERACTION OF PATHOGENS, HOST DEVELOPMENTAL STAGE, AND GENETICS

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Phenotypic plasticity may evolve in response to environmental changes in populations with sufficient genetic variation for the reaction norms. We used the whitefish *Coregonus palaea* to experimentally study the interplay of ecology, genetics, and development on the evolutionary potential of plastic traits. In order to disentangle maternal from paternal contributions, and the likely effects of developmental stage from ecological effects, we sampled a natural population, used gametes for full-factorial *in vitro* fertilizations, raised the resulting offspring singly at controlled conditions, and exposed them at one of several points during development to the opportunistic pathogen, *Pseudomonas fluorescens*. Inoculation with the bacterium increased embryonic mortality and resulted in delayed hatching of smaller, less developed larvae. Vulnerability to infection increased distinctly over embryo development. This change coincided with a shift in the importance of maternal to additive genetic effects on survival. Timing of exposure also affected whether there was genetic variability for trait plasticity, and whether traits were correlated across environments. Our results demonstrate that additive genetic effects on plastic traits and of trait plasticities critically depend on developmental stage and hence on the timing of a stressful event.

D20SY05RT16:33R1

THE EVOLUTION OF ALTERNATIVE DEVELOPMENTAL PATHWAYS IN A BUTTERFLY: LOCAL ADAPTATION IN DEVELOPMENTAL THRESHOLDS AND FOOTPRINTS OF RELAXED SELECTION ON LIFE HISTORY PLASTICITY

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The ability of many temperate insects to have more than one generation per year is due to the plastic induction of alternative developmental pathways - leading to either direct development during summer or diapause development during winter. Typically, the number of generations produced within a population decrease with increasing latitude and the developmental threshold determining which pathway to follow is predicted to show a latitudinal cline due to local adaptation to season length. Consequently, when moving north along a latitudinal cline, populations will ultimately become univoltine and only produce one generation per year, which always enter diapause development. In such locations where only the diapause developmental pathway is expressed, selection on developmental regulation of the direct pathway will be relaxed. I will present results showing how the photoperiodic threshold determining the pathway decision in the butterfly Pararge aegeria is locally adapted to season length. Moreover, I will explore footprints of relaxed selection on the developmental regulation of life history traits and sexual dimorphism when expressed in the direct development pathway. The results suggest that populations experiencing relaxed selection on the direct pathway show less pronounced differences between pathways in several life history phenotypes. In particular, relaxed selection on direct development was associated with a disruption of protandry (earlier emergence of adult males) expressed as sexual dimorphism in larval development time and growth rate. This suggests that relaxed selection on the direct pathway has allowed life history traits to drift towards trait values associated with lower fitness, and that ongoing selection is necessary for upholding this type of "fine tuning" of alternative developmental regulation.

D20SY05RT16:57R1

THE ROLE OF PHENOTYPIC PLASTICITY IN THE RESPONSE OF SPECIES TO ENVIRONMENTAL CHANGE: A RECIPROCAL TRANSPLANTATION OF FIVE GRASSES SPECIES ALONG AN ALTITUDINAL GRADIENT

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Alpine environments are particularly susceptible to environmental changes associated to global warming. The response of species to such environmental changes depends on the relative importance of adaptive variation and phenotypic plasticity. We describe here a reciprocal transplantation experiment of five species across two elevation sets. We measured the response to transplantation and the effect of the surrounding vegetation on survival and biomass as a proxy for plant fitness. Survival and biomass varied according to the elevation of transplantation and to the presence (or absence) of surrounding vegetation, indicating high phenotypic plasticity. On the other hand, although we detected significant differentiation between populations in several species, we found no indication of local adaptation of plants to their population of origin suggesting that phenotypic plasticity is high enough to allow individuals to occupy different environmental conditions. The presence of surrounding vegetation facilitated the survival in stressful conditions while we observed competition for growth at more favorable sites (low elevation). The large response of species in our experiment, together with similar results from other experiments in the same environment, indicates that phenotypic plasticity is an important component of plant adaptation along an altitudinal gradient and is an important component of the response to environmental changes for plants that remain in the same sites as well as an advantage for colonizing new favorable habitats.

D20SY05RT17:45R1

PLASTIC MATING STRATEGIES: TRANSCRIPTOMICS OF MALE RESPONSES TO MATING RIVALS IN DROSOPHILA MELANOGASTER

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Behavioural plasticity is an important strategy that enables animals to respond to short term fluctuations in the environment. The socio-sexual environment can vary rapidly, dictating the level of mating opportunities and competition an individual encounters. Males of many species show plasticity in response to this variation. For example, in *Drosophila melanogaster*, males show a sophisticated and consistent response to the threat of mating competition as signalled by the presence of rivals. Following exposure to a rival male, focal males mate for significantly longer than their counterparts held alone. This results in a significant increase in male fitness in both competitive and non-competitive matings. Males use a complex set of cues to assess the presence of rivals. However, there are also costs, and individuals that maintain responses to rivals throughout life show reduced lifespan and mating success in older life. Plastic responses to rivals are highly flexible and associated with variation in the transfer of ejaculate proteins into females during mating. In this study, we investigated the underlying transcriptomic changes that characterise responses to rivals, using a global mRNAseq approach. We analysed gene expression changes according to the length of exposure to rivals and within different tissue types (the head and thorax versus abdomen). This enabled us to identify highly differentially expressed candidate genes associated with sensing rivals, with temporal changes in behaviour and with manipulating ejaculate content in response to exposure to rivals. These data allow us to investigate the phenotypic role of these transcriptional changes and explore how such a response is regulated. The work is important for understanding how sophisticated and flexible behavioural plasticity can evolve.

D20SY05RT18:09R1

PLASTICITY IN SIGNALLING BEHAVIOUR REFLECTS GENETICS AS WELL AS RELATIVE SIZE IN BURYING BEETLES (*NICROPHORUS VESPILLOIDES*)

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In animals with intra-sexual competition for mates signalling strategies to attract reproductive partners often vary between individuals. How much of this variation is conditional and adopted when an individual is likely to be at a competitive disadvantage and how much is genetically based is a difficult question to address, as there can be strong pleiotropy between the conditions (e.g., social status or size) and the likelihood of adopting a given strategy. To tease apart the contributions of genetics and competitive status of males here we compare the likelihood of adopting a resource-based or satellite signalling behaviour in populations of burying beetles that have experienced 14 generations of artificial bi-directional selection for mating rate. We hypothesized that if signalling behaviour is genetically influenced, males selected for high mating frequency would exhibit more persistent signalling behaviour when on a resource required for reproduction (mouse carcass) than males selected for low mating frequency. Alternatively, if signalling tactics are conditional, we predicted that relative size would influence signalling behaviour more than which selection line males were from. We found that the extent of signalling on a carcass reflected the focal males relative size more than selection history. However, the extent of "sneaking" behaviour (signalling off the carcass) reflected both selection regime and relative size. Thus, we find that genetics and conditional influences make different contributions to alternative mating strategies.

D20SY05RT18:33R1

LOSS OF DIAPAUSE RELATED BEHAVIORAL PLASTICITY DURING NORTHWARDS RANGE EXPANSION IN THE INVASIVE COLORADO POTATO BEETLE LEPTINOTARSA DECEMLINEATA* (SAY) IN EUROPE

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The photoperiod is an important environmental trigger of plastic life-history decisions in many organisms living in seasonal environments. In animals undergoing rapid latitudinal range expansion, a change in the timing of photoperiodic triggering of overwintering behavior is often required for establishment and persistence at novel latitudes. Successful overwintering requires a synchronization of behavior with many physiological traits. While several cases of rapid photoperiodic adaptation have been documented, less attention has been given to flexibility in the overwintering behavior itself and some studies indeed suggest that flexibility in behavior could offset the need for physiological photoperiodic adaptation, through stress avoidance or buffering. This could be one reason behind the success of several invasive species. We studied overwintering behavior and physiology of Colorado potato beetles from three populations in Europe along a latitudinal axis, and show adaptive variation in critical photoperiod (when 50% of adults burrow). However, we also found that burrowing behavior itself is flexible, meaning that beetles can burrow and remain so for prolonged periods even though physiologically unprepared for overwintering. The association between overwintering behavior and underlying physiological traits was stronger in the northern than the southern populations, indicating that selection has reduced variation in burrowing behavior during the invasion progress, possibly due to harsher winters. Our results indicate that flexible behavior alone, without physiological adjustments, is not sufficient for persistence in novel high latitudes, even though it might facilitate range expansion in more benign environments. Thus phenotypic plasticity, in our study the response to the photoperiod by either producing an overwintering or reproducing phenotype is not clear cut, but depends on trait associations between behavior and physiology in a latitude dependent manner.

D20SY05RT18:57R1

TEMPERATURE STRESS OF PRECEDING GENERATIONS MODIFIES THE RESPONSE TO INSECTICIDE STRESS IN THE INVASIVE COLORADO POTATO BEETLE

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Environmental stress is considered to play a significant role in species invasions. As invasive species expand their range to new areas they have to tolerate and adapt to various stressors such as changes in temperature and stress imposed by human action. It has been hypothesized that phenotypic plasticity may facilitate adaptations by providing broad stress tolerance or inducing adaptive responses when organisms face novel environments. Thus far, only few experimental studies have experimentally investigated the short-term responses of organisms to stressors and how these responses can affect their invasion potential. The influence of cross-generational stress effects on species invasions has received even less attention. We studied whether parental temperature stress affects tolerance to insecticide in the insecticide resistant Colorado potato beetle (Leptinotarsa decemlineata) population. We results show that beetles were in general tolerant to stress. The parental temperature stress alone affected beetles positively (increased adult body mass) but it impaired their tolerance to insecticide exposure. In contrast, offspring from the favourable temperature regime showed compensatory weight gain in response to insecticide exposure. Our study show that exposure to stress can involve various responses which can differ in the degree and direction (beneficial, harmful) depending on whether preceding generations have experienced temperature stress. Our study emphasizes the importance of evolution as well as plastic responses in the invasion of species to novel environments.

D21SY05RT11:18R1

INDIVIDUAL VARIATION IN BEHAVIORAL PLASTICITY AND SOCIAL ENVIRONMENTS

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Over the last few decades, it has become evident that between-individual variation in behavior characterize a many animal taxa. There is also increasing evidence that individuals differ in their plastic response to the environment. Various adaptive hypotheses have been suggested, and empirically tested, for why natural selection might favor between individual differences in average behavior and also between individual differences in plasticity. One key hypothesis predicts that differences betweenindividuals in their average behavior, as well as individual differences in behavioral plasticity are favored because social interactions lead to a diverse array of social niches. The social niche hypothesis, predicts that in high competitive environments, there will be select for high behavioral differentiation between individuals and high consistency in behavioral expression (lower plasticity), in order to reduce competition. To empirically test whether patterns of between individual variation (average behavior) and within individual variation (plasticity) support the predictions of the social niche hypothesis, we studied 12 Great tit nest box populations in Southern Germany. To study the patterns of variation between-individuals and within-individuals in the different populations, we quantified aggressiveness four times during their breeding season for 610 great tit breeding attempts in a period of three years (n=2246 tests). The populations differ in average density, but also within populations density varied across years. We used this spatial and temporal variation in social environment characteristics to test whether patterns of variation between and within individuals are indeed a function of social environment characteristics. We further studied the evolutionary consequences of behavioral variation at these two levels by quantifying how the interaction between average behavior, and plastic response to the social environment affects the fitness of individuals.

D21SY05RT11:42R1

PHENOTYPIC PLASTICITY OF SOCIAL BEHAVIOURAL RESPONSE IN A SPATIAL COGNITIVE TASKS

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Phenotypic plasticity enables animals to cope with environmental variations. According to the conditions, animals can use different kind of informations which enable them to adapt their behaviour to the environmental conditions. First, they can use informations from their environment like visuals cues. Second, they can use social informations by observing others individuals' behaviours or interacting with them. Differential use of personal versus social informations may depend on their relative costs in differents conditions. Here we focused on this phenotypic plasticity of the use of information. Using *Drosophila melanogaster* as biological model, we took an interest in the effects of some genetics and social environment variations on the plasticity of informations use during spatial learning task. Based on the existence of natural genetic polymorphism at the foraging gene (known to affect some behaviours), our experiments consisted in analysing variations in the use of informations depending on the genetic nature of one individual or a group and the interaction between each other. In order to manipulate visuals and social cues, we used a device similar to the Morris Water Maze. In this device, Drosophila could use wall visual pattern cues or observation of other individuals' behaviours to find and memorize the location of a cold spot to escape from the hot ground of the arena. The results revealed that genetic variation at the foraging locus is related to variation in the use of social information during spatial learning. While both allelic variants were able to use the different kind of informations during the training trials, a probe trial showed that individuals carrying Sitter allele use more social informations than those carrying Rover allele. These results suggest that rover and sitter individuals don't cope with their environment in the same way but they can switch from one kind of information to another according to the environmental variations.

D21SY05RT14:00R1

EVOLUTION OF DISCRETE PHENOTYPES FROM CONTINUOUS NORMS OF REACTION

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Discrete phenotypic variation often involves threshold expression of a trait with polygenic inheritance. How such discrete polyphenisms evolve starting from continuously varying phenotypes has received little theoretical attention. We model the evolution of sigmoid norms of reaction in response to variation in an underlying trait, or in a continuous environment, to identify conditions for the evolution of discontinuity. For traits with expression depending on a randomly varying underlying factor such as developmental noise, polyphenism is unstable under constant phenotypic selection for two selective peaks, and reaction norm evolution results in a phenotypic distribution concentrated at only one peak. But with frequency-dependent selection between two adaptive peaks, a steep threshold maintaining polyphenism generally evolves. For inducible plastic traits with expression conditioned on an environmental variable that also affects phenotypic selection, the steepness of the evolved reaction norm depends both on the differentiation of the environment in time or space, and on its predictability between development and selection, while bimodality of the environment has little effect per se. Together with recent measurements of genetic variance of threshold steepness, these predictions suggest that quasi-discrete phenotypic variation may often evolve from continuous norms of reactions, rather than being an intrinsic property of development.

D21SY05RT14:24R1

CAUSES AND CONSEQUENCES OF DEVELOPMENTAL PLASTICITY IN DAPHNIA MATURATION

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Maturation is a key life history transition, due to the importance of age and size at maturity in determining fitness. Understanding how maturation phenotypes evolve requires an appreciation of the underlying ontogenetic mechanisms, including the maturation threshold, which determines when an individual 'decides' to mature. Maturation thresholds are poorly understood, and little is known about how phenotypically plastic or genetically variable they are, but the parthenogenetic crustacean Daphnia is the ideal organism in which to study their evolution. Statistically modelling the maturation process shows that the maturation threshold is a developmentally plastic trait in response to variable resource availability, and more closely resembles a process with a rate than a discrete switch. The idea that the threshold is better thought of as a rate than a switch is further supported by gene expression changes during maturation. The maturation threshold also differs between genotypes and species of Daphnia, and clone-specific maternal effects in the development and growth rate interact to produce phenotypically plastic adult phenotypes. Furthermore, experiments studying the fitness consequences of maturation variation showed that *Daphnia magna* genotypes initiating maturation at smaller sizes had a higher intrinsic rate of population increase, but this size did not correlate well with competitive success when five clones were directly competed with each other, suggesting that interactions with other factors were influencing fitness. Maturation thresholds in *Daphnia* do not appear to be based on a single fixed state, but are responsive to environmental variation. The presence of heritable variation and transgenerational effects in these developmentally plastic traits suggests that they have an important role in the evolution of age and size at maturity.

D21SY05RT14:48R1

DISCOVERING THE GENETIC BASIS OF TORPOR IN A CHILEAN MARSUPIAL

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Torpor is the physiologically controlled reduction of metabolic rate and body temperature experienced by small endotherms when facing periods of low temperature and/or food resources. This phenotype is characterized by an almost complete suppression of all expensive physiological processes with the aim of reducing energy expenditure. Nevertheless, some processes continue to operate at lower levels of activity, as they are critical for survival. The high demand of energy required during rewarming, to reach normothermy, represents an important constraint. Torpor and arousal from torpor involves a complex physiological reorganization at different organizational levels, underpinned by changes in genes expression. Accordingly in this study we investigated the reaction norm of (1) gene expression and (2) mitochondrial performance along different stages of torpor bout (deep torpor, arousal and normothermy) in the Chilean marsupial *Thylamys elegans*. More specifically we (1) performed a largescale gene expression screening (RNA-seq) and (2) examined mitochondrial oxygen consumption and different enzymes of the electron transport system associated with torpor in liver. The gene expression profiles revealed a modest level of transcriptional changes along different stages of torpor bout. Functional analysis shows that genes involved in pathways associated to lipid metabolism are increased, whereas those involved in protein biosynthesis and detoxification are decreased during torpor and rewarming. For mitochondrial performance, high level of phenotypic flexibility was observed during the different stages of torpor. Taken together, these findings revealed important metabolic process those are critical during torpor in marsupials.

D21SY05RT15:12R1

A DEVELOPMENTAL SWITCH GENE FOR A FEEDING DIMORPHISM IN PRISTIONCHUS NEMATODES IS COUPLED TO MICRO- AND MACROEVOLUTION OF PLASTICITY

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The mechanistic study of ecologically relevant traits is essential for understanding how ecology and development interact in the evolution of novel phenotypes. The nematode model *Pristionchus pacificus* shows plasticity in its teeth-like feeding structures, which are a novelty that enable predation of other nematodes. The plasticity consists of two discrete forms, stenostomatous (St) and eurystomatous (Eu), the latter bearing a claw-like dorsal tooth and an opposing subventral tooth. To study the genetic basis of the dimorphism, we used forward genetics to isolate Eu-form-defective (eud) mutants. A mutant with dominant alleles, eud-1, is haploinsufficient and was rescued by genetic transformation with a wild-type allele. Extra copies of this X-linked gene also drive highly St males to be Eu, indicating a role for EUD-1 in sexual dimorphism. Overexpression of *eud-1* results in saturation of the Eu form and therefore acts as a dose-dependent master switch for the dimorphism. Further experiments revealed that EUD-1 is not only necessary and sufficient for the mouth-form decision in mutants but also represents a key determinant of micro- and macroevolutionary diversification. A survey of over 100 wild populations of *P. pacificus* gave a population genetics context, revealing natural variation in mouthform phenotypes. Variation correlated with *eud-1* expression, and genetic transformation of highly St strains confirmed the role of EUD-1 as a dimorphism switch. Thus, a gene identified by laboratory genetics can be a key player in natural history. Recent discovery of a sister species to P. pacificus, P. exspectatus, allows tests across species boundaries by the success of hybrid crosses, and transformation by this technique showed maintenance of the EUD-1 switch in macroevolution. By integrating genetics analysis, phylogenetics, and natural history, studies in this system ultimately aim to test the role of phenotypic plasticity in the evolution of novelty.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

D21SY05RT15:45R1

SPATIO-TEMPORAL COMPARTMENTALIZATION OF ECDYSTEROID EFFECTS ON POST-GROWTH PATTERNING

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Developmental plasticity is a phenomenon whereby a single genotype produces distinct phenotypes depending on environmental conditions experienced during development. This process is regulated by changes in endocrine physiology, and has one of its most compelling examples in butterfly wing patterns that differ dramatically across seasons. In the butterfly *Bicyclus anynana*, larvae that develop during the wet season produce adults with conspicuous wing patterns, while those that develop during the dry season produce adults with cryptic patterns. The temperature regulation of the alternative adult phenotypes is mediated by changes in the internal ecdysteroid hormone dynamics. Manipulation of hormone titers copy the effects of temperature. However, little is known about hormonal sensitivities in relation to different external temperatures, to when the manipulations are done in terms of developmental timing, and to how different regions of the same tissue respond. Our aim was to investigate the compartmentalization of the effects of manipulations of systemic hormone levels during pupal development on adult wing patterns. We manipulated external temperatures (representing the natural extremes and an intermediate temperature) and internal levels of 20-hydroxyecdysone (via hormone injections at different developmental time points), and analyzed phenotypic effects on different wings, wing surfaces, color pattern elements, repeated elements of the same type, and groups of cells of the same repeat. Our results show that the effects of hormone manipulations depend on temperature and time point, and is highly compartmentalized in space: with specific epidermal cells responding in specified ways. We also show that this compartmentalization does not reflect compartmentalization of expression of hormone receptor. It remains to be seen whether the spatial compartmentalization of hormone effects is determined upstream or downstream of the binding of the hormone to its receptor.

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MOLECULAR AND GENETIC MECHANISMS UNDERLYING VARIATION IN PHENOTYPIC PLASTICITY IN STICKLEBACKS

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Phenotypic plasticity plays important roles in adaptation to changing environments. However, organisms tend to lose the capacity for phenotypic plasticity under stable environments. Although variation in phenotypic plasticity has been found throughout the animal kingdom, molecular and genetic mechanisms underlying such variation remain elusive. We are addressing this question by using threespine stickleback (Gasterosteus aculeatus), because they have a variety of ecotypes that show different levels of phenotypic plasticity. After the last glacial recession, ancestral marine sticklebacks colonized newly formed freshwater habitats, resulting in extensive phenotypic diversification. In this study, we report that whole transcriptome analysis revealed that marine sticklebacks exhibited significant photoperiodic response of expression levels of thyroid hormone-stimulating hormone beta 2 (TSHß2) gene, whereas such response was lost in freshwater sticklebacks. Loss of TSHß2 response independently occurred in both North American and Japanese freshwater populations. Further analyses of TSHB2 response in F1 hybrids demonstrated that loss of TSHB2 response has different genetic basis between Japanese and North American populations. Genome sequencing further revealed several ecotype-specific SNPs at the cis-regulatory region of TSHß locus in North America freshwater populations. We are currently conducting luciferase-reporter assays in vitro to investigate the cisregulatory mechanisms of TSHB2 response. Furthermore, we are making TALEN-induced knockout sticklebacks to understand the roles of TSH\$2 in vivo. Thus, further studies on the variation in TSH\$2 response will lead to a better understanding of the genetic mechanisms underlying convergent loss of phenotypic plasticity.

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PHENOTYPIC AND GENETIC RESPONSES OF LIFE-HISTORY TRAITS TO PREDICTABLE AND UNPREDICTABLE DEVELOPMENTAL CONDITIONS IN A SEASONAL BUTTERFLY

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Environmental conditions experienced during development are known to influence the phenotype but also the different components of heritability, including genetic variance (VG). Here, phenotypic as well as genetic responses of life-history traits to two different developmental conditions, temperature and food limitation were assessed. The former represents an environment that defines seasonal polyphenism in our study organism, the tropical butterfly Bicyclus anynana, whereas the latter represents a more unpredictable environment. While development time, pupal mass, and resting metabolic rate showed no genotype-by-environment interaction for genetic variation, for thorax ratio and fat percentage the VG increased under the cool temperature, dry season environment. Additionally, for fat percentage, VG increased under food limitation. Hence, the traits most intimately related to the polyphenism in *B. anynana* show the most environmental specific genetic variance as well as some indication of cross-environmental genetic correlations. I will relate these results and the observed phenotypic responses to temperature (season) and food limitation to our recent RNA-Seq analyses on 72 individuals from the same families. We find substantial genetic variation for gene expression variation, affecting 1225 genes significantly (FDR<5%), while seasonal and food conditions affected much fewer genes. Interestingly, we also identified a number of genes whose expression was affected by the interaction between developmental conditions and genetic background, indicating genetic variation for environmental responses.

D21SY05RT16:57R1

NO STRAIN NO GAIN: GENETIC INVESTIGATION OF ADAPTIVE PHENOTYPIC PLASTICITY IN TELEOST JAWS

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Through incorporating environmental signals into ontogenetic pathways, phenotypically plastic species can fine-tune their phenotypes to precisely match local environmental conditions. In spite of its importance in the generation of adaptive phenotypes, the molecular basis of phenotypic plasticity remains poorly characterised. We are establishing a new molecular model for phenotypic plasticity research: the East African cichlid fish, Astatoreochromis alluaudi, which has been the subject of morphological studies of plasticity for 50 years. In response to a hard diet, its Lower Pharyngeal Jaw (LPJ) develops a 'molariform' morphology, with molar-like teeth set in an enlarged, dense jaw, compared with the smaller, finer 'papilliform' morphology which represents the ground state for this species. We performed a common garden experiment where siblings were fed either whole snails (hard diet), or finely minced snails (soft diet) and analysed the resulting morphological and transcriptional phenotypes. Genome-wide transcriptome analysis was performed on the LPJs of molariform and papilliform morphs that resulted from the diet treatments, shedding light on the environmentally sensitive pathways that modulate LPJ morphology. Numerous genes of mechanically responsive pathways (such as fos and jun) were upregulated in the molariform LPJs, indicating that mechanical strain intersects with bone developmental pathways that shape the molariform morphology. Intriguingly we also observe the down-regulation of various inflammatory factors and redox pathway members, suggesting that microenvironmental changes within the LPJ medullary cavity may alter cellular differentiation and proliferation. Through identifying numerous pathways involved with plasticity in the LPJ of A. alluaudi, our research opens the door to assess the role of phenotypic plasticity in generating morphological novelties amongst the explosive radiation of East African cichlid fishes.

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IS PREDATOR-INDUCED PHENOTYPIC PLASTICITY REGULATED BY DNA-METHYLATION IN TADPOLES?

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Phenotypic plasticity is the ability of a genotype to modify its phenotype in response to reliable environmental cues. The knowledge of the mechanisms involved in plasticity is the key to understand its ecological, developmental and evolutionary consequences. In the last decade, epigenetic changes have been pointed out as explanations for several phenotypically plastic changes. In particular, DNA methylation has been linked to phenotypic plasticity and evidence is quickly accumulating across different taxa. Here we tested if the predator-induced phenotype observed in many amphibian larvae was the consequence of changes in gene expression derived from changes in the pattern of DNA methylation. Tadpoles produce anti-predator phenotypes that include a conspicuous increase in relative tail depth and a reduction of activity rate. To test if these plastic responses are epigenetically regulated, we exposed *Pelobates cultripes* tadpoles to the presence or absence of chemical cues from predators, while previously treating or not tadpoles with 5-azacytidine, a DNA methylation inhibitor, in a 2x2 factorial experiment. Our experimental treatments were: control (clean water), predator cues (dragonfly nymphs, Anax imperator), 5-azacytidine, and 5-azacytidine plus predator cues. Tadpoles exposed to predator cues showed the expected antipredator morphology, whereas tadpoles exposed to predator cues but earlier treated with 5-azacytidine showed an intermediate phenotype between the control and predator exposed tadpoles. They also showed a substantial decrease in activity rate as compared to control tadpoles and even to predator-exposed tadpoles. We are currently undertaking a molecular analysis with methylation-sensitive amplified length polymorphism (MSAPs), to quantify the changes in epigenetic marks. In view of these results, it seems like epigenetic regulation of predator-induced phenotypic changes in *Pelobates tadpoles* may be at least partially driven by DNA-methylation.

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DIFFERENTIAL GENE EXPRESSION IN A NON-MODEL FISH SPECIES WITH ALTERNATIVE MATING TACTICS

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Social dominance is important for the reproductive success of males in many species. In fish with external fertilization, it is not as apparent which traits are necessary to become dominant or territorial and what benefits the socially dominant individual. In the black-faced blenny (Tripterygion delaisi) during the reproductive season, some males change color and invest in nest making and defense, whereas sneaker males do not change color and 'sneak' reproductions when females lay their eggs. Using RNAseq, we profiled differential gene expression between the brains of territorial males, sneaker males, and females to study the molecular signatures of male dimorphism. Despite several studies reporting high levels of genetic differentiation between sexes, we found that more genes were differentially expressed between the two male phenotypes than between males and females. This suggests that phenotypic plasticity is a more important factor in differential gene expression than sexual dimorphism during the reproductive period. For the dominant male, expression was higher in genes mainly related to cytoskeletal rearrangement indicating the drastic change in behavior and phenotype. We also identified novel genes which are differentially expressed in the brain tissue between the two male mating types in Tripterygion delaisi, which can be further investigated in other fish species with similar mating tactics.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

D21SY05RT18:33R1

SEX ALLOCATION PLASTICITY ON A TRANSCRIPTOME SCALE

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Predicting an organism's optimal sex allocation has long been a central concern of evolutionary biology research. In simultaneous hermaphrodites, the question concerns the balance of investment into the male versus the female sex function. Theory predicts that individuals should bias investment more towards the male function as the number of available mates increases, and this is indeed a welldocumented phenotypically plastic response in the free-living flatworm Macrostomum lignano. However, the details of how sex allocation plasticity is achieved at a molecular level are currently unknown. To address this, we performed an RNA-Seq experiment on worms raised in different social environments (i.e. in isolation, in pairs or in octets), known to lead to different optimal sex allocations, and investigate how these worms respond on a transcriptome scale. We demonstrate that up to 10% of all known transcripts are differentially expressed between different social environments, with this figure rising to >30% for gonad-specific genes that are presumably directly involved in the switch away from oogenesis towards spermatogenesis under increased mating group size. As predicted, most differentially expressed testis-specific genes are upregulated in larger groups, and most differentially expressed ovary-specific genes are downregulated in larger groups (although there are some notable exceptions to this general pattern). Moreover, we identify many tail-specific genes that are upregulated in larger groups, many of which are prostate-specific genes involved in seminal fluid production, a previously unquantified aspect of male allocation. Our data provide a rich repertoire of candidate genes for functional characterization in the context of the control of gametogenesis and sex allocation (which we can now investigate using RNAi and other tools available in this system), and offer broad insights into the molecular underpinnings of phenotypic plasticity.

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TRANSCRIPTOME ANALYSIS OF PREDATOR- AND PREY-INDUCED PHENOTYPIC PLASTICITY IN THE HOKKAIDO SALAMANDER (HYNOBIUS RETARDATUS)

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Larvae of the Hokkaido salamander (Hynobius retardatus) exhibit two distinct morphs, "attack morphs" and "defense morphs", as a result of inducible phenotypic response to preys and predators, respectively. The existence of preys, Rana pirica tadpoles, leads to the induction of attack morphs, which have broad heads likely suitable for catching tadpoles. The existence of predators, dragonfly larvae (Aeshna nigroflava), induces them to become defense morphs, which have enlarged external gills and high tails suitable for avoiding the predatory attacks. However, molecular mechanisms underlying this phenotypic plasticity have yet to be elucidated. To reveal the developmental and physiological mechanisms of this phenotypic plasticity, we carried out de novo transcriptome analysis of the Hokkaido salamanders. First, we collected eggs in the wild and then hatched them in laboratory. The larvae were exposed to either predators or preys to induce different morphs. Morph induction was completed about 7 days after the start of exposure. RNAs were extracted from 4 tissues (brain, head, external gill and tail) and 3 time points (0 hour, 12 hours, and 7 days after the exposure onset) and sequenced. Obtained reads and contigs of treatment samples were compared with those of control samples (i.e. no exposure) to identify differentially expressed genes. Approximately 2,000 genes were identified as differentially expressed genes at each time point and in each tissue. These genes include hormone related functional genes, such as COL2A1, HBA2, and NR2C1. RNA processing/splicing was found as an enriched functions in the Gene Ontology database, suggesting that the phenotypic plasticity of Hokkaido salamanders may be linked to regulation of alternative splicing.

POSTERS

REACTION NORMS FOR SIZE AND TIME AT MATURITY IN INSECTS: PHENOTYPIC PLASTICITY VERSUS CONSTRAINTS

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Explaining the shape of reaction norms for size and time at maturation is one of the major questions of the theory of life history evolution. Classical models predict negative correlation between final body size and development time across different environmental conditions: it takes little time to grow large when conditions are good, whereas it takes longer to grow but size remains smaller when conditions are poor. Nevertheless, recent theoretical developments suggest that, under specific circumstances, a wide array of possible shapes of the size vs. time reaction norms could be optimal, including reaction norms with a positive slope. Empirical evaluation of these predictions has remained insufficient so far. Here we present a review of nearly 200 published case studies on insects where the shape of the time vs. size reaction norm could be derived. We found that the "classical" negative slopes clearly predominate when the environmental gradient is based on resource quality but there are meaningful exceptions. Additionally, we report results of original case studies on reaction norms of body size determination in several lepidopteran species. We point at similarities of unrelated species which could indicate the presence of constraints stemming from the evolutionarily conservative developmental physiology of insect larvae, and caution against a pan-adaptationistic interpretation of respective reaction norms.

FEMALES PLAY IT COOL: PLASTICITY OF POLYANDRY IN NORTH AMERICAN DROSOPHILA

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Some females mate once in their lives, others with a large number of males, and the reasons for this intraspecific variation in polyandry are not clear. Temperature is a potentially important environmental variable that can influence polyandry in populations occupying different environments. This is due to the fundamental relationship between temperature and biological reactions governing growth and development in juveniles and physiological stress in adults. Since polyandry plays a key role in regulating population fitness, it is essential to address how a major environmental variable affects plasticity of polyandry. We examined plasticity of polyandry over four temperature environments using 26 genotypes of the fruit fly Drosophila pseudoobscura from locations across the USA. Genetic variation proved the strongest driver of intraspecific patterns of polyandry. We also found that females were more polyandrous when exposed to cooler temperatures during mating trials than when exposed to warmer temperatures. Despite this, we found no evidence for genetic variation in plasticity of polyandry as all genotypes responded to temperature in a comparable way. Temperature experienced by females as larvae had no effect on subsequent adult mating behaviour. These results clearly indicate that only the current temperature experienced by females promotes plasticity of polyandry and that the major explanation behind intraspecific variation in mating behaviour is genetic. This suggests that polyandry is maintained under localised selection and challenges the notion that fitness benefits of polyandry are temperature-dependent. This is an important step towards determining how behavioural plasticity contributes towards overall population fitness, and ultimately how the relationship between genetic and environmental heterogeneity can influence species distribution patterns.

BETWEEN-HOST PHYLOGENETIC DISTANCE AND PERFORMANCE OF HAEMATOPHAGOUS ECTOPARASITES

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Parasites vary in their abundance among host species. The host used by the majority of parasite individuals is considered the principal host, while the remaining host species are referred to as auxiliary hosts. Variation in parasite abundance among auxiliary hosts reflects the degree of phylogenetic proximity between the principal host and the auxiliary hosts it used. The mechanism underlying this pattern is expected to be related to differential performance (feeding and reproductive) of parasites in auxiliary hosts that differ in their phylogenetic distance from the principal host. We tested this hypothesis using fleas parasitic on small mammals. Although feeding performance (blood meal size, energy expenditure for digestion and time of digestion) of parasites differed among different hosts, (1) they did not always perform better on a principal host than on an auxiliary host; and (2) their performance on an auxiliary host was not negatively correlated with phylogenetic distance of this host from the principal host. In accordance with our hypothesis, reproductive performance of parasites (egg and/or new imago production) in an auxiliary host decreased significantly with an increase in phylogenetic distance between an auxiliary and the principal host. However, this was true only for auxiliary hosts belonging to the same family as the principal host. One of the proximate causes for lower reproductive performance and subsequent lower abundance of parasites on auxiliary hosts appeared to be the higher energy cost of egg production in the latter. However, in some parasite species, lower offspring number in an auxiliary host was compensated to some extent by offspring size, although this compensation might also compromise parasites' future reproduction via decreased survival. In other words, reproductive strategy implied during exploitation of low profitable (i.e., auxiliary) hosts may differ between parasite species.

STABILITY OF SEX EXPRESSION IN THE GYNOMONOECIOUS-GYNODIOECIOUS PLANT SILENE ITALICA

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Gynomonoecious individuals are individuals that bear both perfect and female flowers. There are sometimes found in gynodioecious species, namely together with female and hermaphroditic individuals. This kind of reproductive system allows variations of floral sex ratio within individuals and could thus be interpreted as a mean to vary sex allocation. Change in sex according to the internal conditions of the plant (e.g., size) or to environmental conditions can be adaptive if male and female functions respond differently to these conditions and if these conditions change though time. Sex instability can also be non-adaptive and a consequence of sex determination mechanisms. In Silene italica L., a gynodioecious species with gynomonoecious individuals, we examined how the sex of individuals, defined as their proportion female flowers, varied with age, size or environment. We also checked if the female flowers of gynomonoecious individuals occurred at the beginning of the season or on specific positions within the plant. These data were use to test if sex change in this species fits the "adaptive hypothesis" or is rather in accordance with the "developmental noise hypothesis".

GENE EXPRESSION DIFFERENCES AMONG CASTES AND LIFE STAGES IN THE ANT FORMICA EXSECTA, A TRANSCRIPTOME STUDY

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Understanding how the interaction of genotypes and environment produces distinct phenotypes from similar sets of genes is a crucial topic in evolutionary biology. The queen and worker castes of social insects are a striking example of such polyphenism. In social Hymenoptera both the workers and reproductive queens are females, and develop from diploid eggs. In ants, caste polyphenism is characterized by extensive phenotypic differences, with queens that specialise in reproduction being usually much larger than workers who carry out brood care, foraging, nest maintenance and defence. Because the phenotypically distinct individuals develop from the same set of genes, these morphological, physiological and behavioural differences found between worker and queen must be due to variation in gene expression. However, little is known about qualitative and quantative variation in caste specific gene expression patterns across life stages. Because expression patterns of genes partly determine the strength of natural selection on them, separation of gene expression differences in developing and adult individual is crucial for understanding both the causes and consequences of caste differences. Specifically, it is crucial for strength of natural selection whether there are genes that are exclusive to one caste, or whether the same genes are expressed in both castes, but at different points in time. We used *Formica exsecta*, a monogynous ant species, as our model species for this study. We assessed queen and worker gene expression levels at three different life stages (pupae, emerging adult and adult) by sequencing their transcriptome. Samples were collected in Finland during Spring 2011 and two sets of cDNA library were made. As phenotypic plasticity mediated by gene expression differences is the foundation of social insect evolution, we believe that this novel study will bring further knowledge on understanding the molecular basics of phenotypic evolution.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

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MODELING PLASTIC AND CANALIZED PHENOTYPIC EXPRESSION BY THE PROCESS OF GENETIC ACCOMMODATION

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The evolution of polyphenism, in which a single genotype produces alternative discrete phenotypes in response to environmental cues, and the evolution of a novel canalized phenotype, in which a genotype produces a novel, invariant phenotype in spite of environmental differences, represent different end points in the process of genetic accommodation. Genetic accommodation is hypothesized to begin when heritable variation in developmental switch points and environmental variation conspire to trigger the expression of a novel phenotype, thus exposing the underlying genetic variation to selection. We model genetic accommodation using a threshold model. Using simulations, we illustrate the model by application to the evolution of both the environmentally induced expression of a novel defensive phenotype and the evolution of the canalized expression of a defensive phenotype. We show that, given the appropriate ecological and genetic conditions, genetic accommodation can occur rapidly, resulting in the evolution of either polyphenism or a novel canalized phenotype.

GENETIC AND PHENOTYPIC VARIATION IN DEVELOPMENTAL RATE ACROSS GEOGRAPHIC REGIONS: A PHOTOPERIOD DRIVEN LATITUDE COMPENSATING MECHANISM?

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Genetic and phenotypic variation in developmental and growth rates along latitudinal gradients may benefit our understanding of the evolution of latitudinal compensating mechanisms. I explored compensatory developmental mecha-nisms with respect to photoperiod in central, northern and northernmost peripheral populations of a damselfly Lestes sponsa. In addition, genetic variance in developmental and growth rate was evaluated across the populations. L. sponsa is strictly univoltine with egg overwintering throughout its geographic distribution. The role of photoperiod on the expression of larval development was evaluated under controlled laboratory conditions. Larvae from each region were grown in both high and low latitude photoperiod. All three populations shortened development time and accelerated growth as a response to high latitude photoperiod (phenotypic plasticity present). The slopes of reaction norms differed between populations. There was a genetic differentiation between populations and this resulted in a genotype-by-environment interaction in development time and growth rate. Genetic variance (Vg) in development time was significant, however it did not differ across populations. Vg in growth rate was non-significant. Reaction norms in development time and growth rate also showed non-significant Vg. Results supported the presence of countergradient variation in development time and growth rate as seasonally more time stressed populations took shorter time for development and grew faster than less time stressed populations. Slopes of reaction norms indicated that the latitude compensating mechanism was mediated by photoperiod. While growth rate seems to be genetically constrained for further evolution, development time has a potential to evolve, though, at a similar rate across study regions. Reaction norms seem to be canalized with respect to photoperiod.

ECO-EVOLUTIONARY DYNAMICS OF FRESHWATER WATER FISH COMMUNITIES IN POST GLACIAL LAKES

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The processes facilitating the diversification of life on earth have intrigued scientists for centuries. Inter-species competition for resources and optimal utilisation of niche space are thought to be large contributors to diversification. Resource polymorphism is a common consequence of this process – where specialisation on particular resources results in diversification within a species. This has been demonstrated to be a common phenomenon in post-glacial lakes, with fish populations typically specialising on either littoral or pelagic resources. Here we investigate the environmental, community and population-level factors contributing to the degree of resource polymorphism observed in two common and phenotypically plastic freshwater fish, perch (*Perca fluviatilis*) and roach (*Rutilus rutilus*). We find that the potential for niche expansion and release from inter-species competition are important variables leading to the presence of resource polymorphism. In addition, we demonstrate that the phenotypic plasticity observed in these species is facilitated by a lack of assortative mating, and fluctuating selection due to population instability over time, particularly for perch. This study system provides support for the importance of phenotypic plasticity in diversification and the interaction of ecological and evolutionary processes.

ADULT CONSPECIFIC CUES AFFECT MOLTING RATE, SURVIVAL AND CLAW MORPHOLOGY OF EARLY RECRUITS OF THE SHORE CRAB CARCINUS MAENAS

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Besides signaling adequate benthic habitat, conspecific cues often shorten development time to metamorphosis and affect both survival and growth of early juvenile stages. However, aggregation of juvenile cohorts in preferred habitat, usually biogenic substrates holding intricate physical structure and high food supply, may lead to strong intraspecific competition and cannibalistic interactions. Using a simple laboratory experiment and the crab species Carcinus maenas as a biological model, we investigated the effects of cues released by adults on intermolt time, growth, and survival of conspecific megalop larvae and juveniles. Using geometric morphometric analyses, we also compared the size and shape of the carapace and claw of stimulated (St) and control (C) juveniles at both the 1st (J1) and 5th (J5) benthic stages. Results obtained showed that conspecific cues can reduce significantly intermoult time and survival, but these differences are restricted to some specific stages. Neither the size nor the increment at molt differed between treatments. There were no differences of carapace characteristics, but conspecific cues affected claw size and shape of J1 and J5 individuals, respectively. For J1 crabs, claws of St individuals were larger than those of C ones, showing an initial size effect. In the case of J5 juveniles, there were no size differences but evident morphological differences suggest that St crabs bear stronger chelae. By the J5 stage, both St and C juveniles exhibit initial heterochely which precedes the prevailing adult pattern. In spite of reducing survival rate, we conclude that overall effects of conspecific cues are positive. Stimulated juveniles may attain a size-refuge from cannibalism earlier than C individuals, and are likely to more efficiently use valuable feeding resources demanding crushing power, such as mollusk prey, in habitat patches characterized by high density of benthic consumers where competitive interactions are very likely.

THE EVOLUTION OF POLLINATOR EMERGENCE TIMING: MODELING THE INFLUENCE OF ENVIRONMENTAL VARIANCE, INTRASPECIFIC COMPETITION, AND LONGEVITY

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The timing of interannual phenologies like flowering in plants or emergence in insects fundamentally effects the fitness of individuals. Particularly in times of globally changing climatic conditions a deeper understanding of the evolutionary mechanisms shaping phenologies is a prerequisite for scientifically based risk assessment. We analyze the evolution of emergence timing in a population of pollinators. Specifically, we are interested in the influence of longevity, intra-specific competition, and inter-annual fluctuations in season length on the temporal distribution of emergence. In our model we assume that individual phenotypes are characterized by a time of emergence that is under stabilizing selection with respect to an optimal value that fluctuates between years. At the genotypic level individuals are characterized by their offspring distribution with heritable genetic variation for the mean and variance. We use an analytical model and individual-based simulations to show that for short lived organisms intraspecific competition promotes an ideal free distribution of emergence times whereas bet-hedging in a variable environment can lead both to diverse phenotypes due to risk spreading and to a small range of phenotypes by risk-avoiding strategies. The different selective forces thus lead to a compromise between synchronising with the environment and increasing the variability in timing, dependent of the external circumstances and stimuli. It is particularly interesting to note that the observed evolution of genetically distinct emergence times may cause assortative mating and form a basis for sympatric speciation.

LEARNED PREDATOR RECOGNITION ALLOWS ACTIVATION OF DEFENSIVE BEHAVIOUR AND ENHANCES SURVIVAL AGAINST ALIEN PREDATORS

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Alien predators are one of the major causes of rapid decline and extinction of native species, because they often create novel ecological contexts in which the antipredatory responses of native organisms are no longer adaptive. The red swamp crayfish, Procambarus clarkii, is a harmful invasive species in aquatic systems worldwide that is causing great ecological impact on native amphibian populations through intense predation of eggs and tadpoles. Larval amphibians are often capable of innately responding to the presence of chemical cues from local predators through changes in morphology and behaviour. Nonetheless, naïve tadpoles are often incapable of recognising alien predators with whom they have no shared evolutionary history. For this reason, amphibian species are especially vulnerable to the introduction of new predators. However, given enough time, native populations might acquire, by means of rapid adaptation or the evolution of phenotypic plasticity, the ability to escape from alien predators. Here we show that naïve tadpoles of the western spadefoot toad, Pelobates cultripes, cannot innately recognise water-borne cues from P. clarkii. Nevertheless, we also show that P. cultripes tadpoles can learn to recognise the cues of this alien predator as a threat when they are exposed to predator cues combined with conspecific alarm cues. Furthermore, we show that tadpoles that learned to recognise the new predator experienced higher survival during staged predation trials with invasive crayfish. This cognitive ability of tadpoles might be critical for amphibian populations to trigger inducible defences against alien predators, thus tempering the immediate impact of invasions through behavioural plasticity and persist long enough for genetic variants to appear and respond to selection.

TEMPERATURE DEPENDENT GENE EXPRESSION AND THE INVASION OF NEW THERMAL ENVIRONMENTS IN COTTUS

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Phenotypic plasticity is considered to play an important role as a source of phenotypic novelty and changes in plastic traits may contribute to evolutionary change. Here, we test if the modification of preexisting plastic responses contributed to the acclimatization and adaptation to different temperature regimes in freshwater sculpins of the genus *Cottus*. This is relevant to ectothermic fishes because water temperature is a fundamental ecological parameter that structures ecosystems along a river and critically determines physiological processes. While *Cottus* are typically known as stenotopic inhabitants of cold water bodies, a young invasive lineage of hybrid origin is currently colonizing summer-warm habitats. We have thus analyzed gene expression responses to changes in water temperature. Temperature specific expression profiles of four populations of the ancestral species and one population of the invasive lineage were assessed over a temperature range from 14°C to 25°C. This range was chosen to reflect a possible range of summer conditions in the originally occupied and newly invaded habitats. Contrary to our expectation that invasive *Cottus* show a marked response to raised water temperatures, the clearest expression profile differentiation between parent species and invasive *Cottus* was observed at temperatures < 21°C that are common in both habitats. Comparative analysis of population specific gene expression changes over temperature revealed plastic responses that are shared by all Cottus as well as changes in thermal plasticity between the ancestral species and the invasive lineage. In particular the increased plasticity of a mitochondrion related phenotype in the invasive lineage contributes to the functional differentiation from its ancestors. Our results demonstrate that an increase in plasticity of an already existing plastic phenotype played a role in phenotypic differentiation and diversification in *Cottus*.

HOW MATERNAL CALORIC RESTRICTION DURING PREGNANCY AFFECTS APPETITE OF THE OFFSPRING IN LABORATORY MAMMALS: A META-ANALYSIS

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Development of the mammalian foetus is strongly affected by maternal nutrition. Maternal undernutrition can result in significant alterations to postnatal phenotypes. Such alterations can be expressed, for example, as changes in body size and behavioural differences. Altered feeding behaviour in the offspring of food-deprived mothers may underpin increased weight gain and fat accumulation leading to obesity and other adverse long-term health effects. We aimed to explore the link between maternal diet during pregnancy and an offspring appetite via meta-analysis using experimental data from laboratory mammals.. We collected 89 effect sizes from 35 studies, together with relevant moderators. Initial analysis revealed higher food intake per unit of body mass in the offspring of the calorically restricted mothers than in the offspring of the control mothers. Surprisingly, the level of maternal caloric restriction was not related to the magnitude of changes in offspring appetite. Instead, it was the proportion of protein in the maternal diet that influenced the relative food intake of the offspring. This effect appeared to be female-specific, suggesting that female offspring are more sensitive than males to maternal diet composition.

ROLE OF BEHAVIOR AND PHENOTYPIC PLASTICITY IN THERMAL STRATEGIES

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Ectotherms cope with the heterogeneity of a thermal environment using the unique combination of behavioral thermoregulation, thermally-induced plasticity, and evolutionary adaptation. Because of its immediate and reversible response, behavioral thermoregulation has been considered as a filter reducing the variation in body temperatures, and thus the selection pressure on thermal physiology traits. I examined this issue using a non-traditional model in thermal biology, the Alpine newt. Results showed that newts combine various behavioral and physiological components in their thermal strategy, proving their suitability for testing predictions of current theories. Newt response to changing thermal conditions represents not only behavioral adjustments but its combination with thermally-induced plasticity. The relative buffering efficacy of both components is determined by their costs under a given combination of biotic and abiotic factors. These facts should be incorporated into future co-adaptation models of thermal biology.

RATION DEPENDENT ALLOCATION TO GROWTH AND REPRODUCTION IN AN ANNUAL KILLIFISH

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Animals with indeterminate growth experience a life history trade-off in resource allocation between reproduction and growth throughout their lives. Adaptations to erratic conditions of temporal pools in African savannas make annual killifish Nothobranchius furzeri (Cyprinodontiformes) a species with an extremely rapid growth. We tested hypothesis that their resource allocation is also very plastic. Specifically, we studied their ability to track resource abundance by allocation of resources between the growth and reproduction. We experimentally manipulated the diet ration at six treatment groups. Two groups received the same ration throughout the experiment (low-low, high-high). The ration of other two groups was switched in the middle of the experiment (low-high, high-low). Finally, another two groups (low-†, high-†) were sacrificed during the ration switch to enable control for size and fecundity with respect to age and size differences among groups. Female size and fecundity were significantly different between the two rations already in the first part of the experiment and remained different throughout the experiment. In the treatments with the switched ration, growth cessation (high to low ration) and growth acceleration (low to high) were observed. The low-high treatment fish fully compensated the size difference of fish fed high ration throughout the experiment and no decrease in fecundity as a cost of compensatory growth was detected. Overall, only current ration affected female allocation to reproduction, the initial ration had no effect. This demonstrates that growth rate and fecundity of *N. furzeri* females rapidly track changes in resource availability. A decreased growth rate when resource availability is low can be readily compensated when conditions are improved, with no long-term effect on their reproductive potential. Such adaptation is advantageous in the erratic environments with highly variable level of resource availability and competition.

PLASTICITY AND SALMON GROWTH: GENETIC DETERMINISM VERSUS SOCIAL COMPETITION

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Individual growth rate in salmon is highly plastic with the literature reporting strong effects of both genetic and environmental factors. One environmental factor with documented effects is social environment, with large dominant fish typically reducing the growth of smaller subordinates. At the same time, others report on compensatory or catch-up growth with small fish being able to grow faster than normal after falling behind in size due to starvation. I examined the growth responses of starved Atlantic salmon of various sizes when reared together with continuously fed fish under socially stable conditions. I found that the starved fish, that lost growth due to starvation, were able to grow faster than now same-sized continuously fed individuals, but were not growing faster than previously (i.e. prior to starvation) same-sized individuals. The relative size among starved fish (i.e. both small and large starved fish) did not influence their ability to grow faster than their similar-sized fed conspecifics. This indicates that in a stable environment, a small fish is small not because it is outcompeted by larger individuals, but likely because it has a weaker genetic drive acting through the production of growth promoting hormones and peptides.

FITNESS EFFECTS OF WARNING SIGNALS THROUGH DIFFERENT LIFE-STAGES AND PHENOTYPES

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Many plants and animals advertise their unpalatability through warning signals in the form of colour and shape. A trade-off is inevitably faced to either allocate resources to signal efficiency or to other processes such as thermoregulation (i.e. melanin production). In the case where organisms undergo different phenotypes throughout their lifespan, such as Lepidopterans, it is unclear if allocating resources to warning signals in one phenotype can transfer fitness advantages to the next phenotype. Here we address this question by rearing full-sib tiger moth larvae (*Parasemia plantaginis*) in high and low temperature conditions and follow their warning signal development until adulthood. Subsequently, we tested for fitness differences as measured by survival and adult heating and metabolic rates. Our analyses showed that larvae reared at low temperatures had higher survival rate and decreased their signal size along its development, producing more melanised body segments. However, adults reared in higher temperatures had higher amount of melanin in their thorax. No clear differences were observed in the amount of melanin in the fore and hind wings between treatments. Adults reared at high temperature had a faster heating rate. On the other hand, adults reared at low temperature needed longer heating time, but had higher body temperature when flying was engaged. No differences were found between metabolic rates between adults of both treatments. Overall, our results suggest that allocating resources to maximise fitness during one life phase or phenotypic stage, does not necessarily translates into higher fitness in the following stages.

JACK OF ALL TRAITS, ADAPTED TO ALL: THE HIGHLY PLASTIC FRESHWATER SNAIL RADIX BALTHICA ALSO SHOW PLASTICITY IN MANTLE PIGMENTATION

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By having phenotypically plastic traits, such as morphology, behaviour and life history, many organisms optimise their fitness in response to fluctuating threats. Freshwater snails with translucent shells, e.g. snails from the Radix genus, differ considerably in their mantle pigmentation patterns, with snails from the same water body ranging from completely dark pigmented to only a few dark spots. These pigmentation differences have previously been suggested to be genetically fixed, but we suggest that this polymorphism is due to phenotypic plasticity in response to a fluctuating environment. Hence, we here aimed at assessing if common stressors, including ultraviolet radiation (UVR) and predation, induce a plastic response in mantle pigmentation patterns of Radix balthica. We show, in contrast to previous studies, that snails are plastic in their expression of mantle pigmentation in response to changes in UVR and predator threats, i.e. differences among species or populations are not genetically fixed. When exposed to cues from visually hunting fish, R. balthica increased the proportion of their dark pigmentation but without compromising the complexity of these pigmented spots, suggesting a crypsis strategy. Snails increased their pigmentation even further in response to UVR, but this also lead to reduced complexity of the patterns. Furthermore, when exposed to UVR and fish, snails responded in the same way as in the UVR treatment, suggesting a trade-off between photoprotection and crypsis.

PATHOGEN-INDUCED HATCHING AND POPULATION-SPECIFIC LIFE-HISTORY RESPONSE TO WATERBORNE CUES IN BROWN TROUT (SALMO TRUTTA)

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Hatching is an important niche shift, and embryos in a wide range of taxa can either accelerate or delay this life-history switch in order to avoid stage-specific risks. Such behavior can occur in response to stress itself and to chemical cues that allow anticipation of stress. We studied the genetic organization of this phenotypic plasticity and tested whether there are differences among populations and across environments in order to learn more about the evolutionary potential of stress-induced hatching. As a study species, we chose the brown trout (*Salmo trutta*; Salmonidae). Gametes were collected from five natural populations (within one river network) and used for full-factorial in vitro fertilizations. The resulting embryos were either directly infected with *Pseudomonas fluorescens* or were exposed to waterborne cues from *P. fluorescens*-infected conspecifics. We found that direct inoculation with *P. fluorescens* increased embryonic mortality and induced hatching in all host populations. Exposure to waterborne cues revealed population-specific responses. We found significant additive genetic variation for hatching time, and genetic variation in trait plasticity. In conclusion, hatching is induced in response to infection and can be affected by waterborne cues of infection, but populations and families differ in their reaction to the latter.

PHENOTYPIC ADJUSTMENTS AND PROPHYLAXIS IN ANTICARSIA GEMMATALIS LARVAE ARE TRIGGERED BY THE PRESENCE OF CONSPECIFICS

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Defence from parasites and pathogens involves a cost. Thus, it is expected that organisms use this only at high population densities, where the risk of pathogen transmission may be high, as proposed by the "density-dependent prophylaxis" (DDP) hypothesis. These predictions have been tested in a wide range of insects, both in comparative and experimental studies. We think it pertinent to consider a continuum between solitarious and gregarious living insects, wherein: (1) solitarious insects are those that are constitutively solitary and do not express any phenotypic plasticity, (2) the middle of the continuum is represented by insects that are subject to fluctuations in local density and show a range of facultative and plastic changes; and (3) constitutively gregarious forms live gregariously and show the gregarious phenotype even in the absence of crowding stimuli. We aimed to chart some of the intermediary continuum with an insect that presents solitarious aspects, but that is subject to fluctuations in density. Thus, Anticarsia gemmatalis (Lepidoptera: Noctuidae) larvae reared at higher densities showed changes in coloration, a greater degree of encapsulation, had higher hemocyte densities and were more resistant to Baculovirus anticarsia, but not to Bacillus thuringiensis. Meanwhile, with increased rearing density there was reduced capsule melanization. Hemocyte density was the only variable that did not vary according to larval phenotype. The observed responses were not a continuous function of larval density, but an all-or-nothing response to the presence of a conspecific. As *A. gemmatalis* is not known for gregarious living, yet shows these density-dependent changes, it thus seems that this plastic phenotypic adjustment may be a broader phenomenon than previously thought

HABITAT USE AND BODY SHAPE EVOLUTION IN ALL AUSTRALIAN FROGS

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The body shape of anurans is predicted to differ among species for functional reasons and in relation to environmental niche. We quantified morphological differences in shape and size among 239 species of the three main Australian frog families (Myobatrachidae, Hylidae and Microhylidae, all genera and more than 98% of the species represented) in order to test the prediction that habitat type predicts morphological variation. We tested this prediction at two levels – across all genera and within the most species-rich genera. Thirty-four external measurements were taken on 1184 museum specimens. Data for seven key environmental variables relevant to anurans also was assembled for all Australiandistributed species based on species' distributions and 10x10 km2 absence/presence locality records. All three Australian frog families showed high diversity in adult body size, ranging from minute (11 mm in microhylids, 15 mm in myobatrachids and 15 mm in hylids) to large species (45 mm in microhylids, 92 mm in myobatrachids and 104 mm in hylids). They also displayed conspicuous differences in body shape, particularly in relative limb length. The environmental niche defined three different groups: 1) species in wet habitats with relatively long limbs, 2) species in arid environments with relatively short-limbs (many of which are forward or backward burrowers), and 3) habitat generalist species with a conservative body shape. These patterns were not repeated within the most species-rich genera: each displayed a highly conservative anuran body shape yet individual species within each genus are distributed across the full spectrum of Australian environments. The patterns of variation likely reflect both phylogenetic history and adaptive pressure imposed by climate and habitats related to the life history of each species.

THE ROLE OF LOCAL ADAPTATION AND PHENOTYPIC PLASTICITY IN THE POPULATION ESTABLISHMENT AND DIVERGENCE OF A FRESHWATER PROTIST

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Most microalgal species consist like all evolutionary higher organisms of many genetically distinct populations, which inhabit a large variety of habitats. The divergence of different populations is in conflict with the high dispersal ability and assumed continuous gene flow of microorganisms. As many recent publications emphasize the enormous intraspecific diversity in microalgae, this study aims at understanding the phenotypic diversity that it represents. Local adaptation might act as a dispersal barrier and play a role in population divergence. We studied phenotypic plasticity and potential of local adaptation in the unicellular, nuisance freshwater microalgae Gonyostomum semen. This species originates from very acid, brown water swamp lakes, but after extensive spreading in Scandinavia over the last decades it can be found also in clear water lakes with neutral pH. Such a fast invasion together with successful establishment as several genetically distinct populations in so many different habitats requires extensive plasticity and high ability for adaptation. In the present study we performed controlled laboratory experiments to test whether strains from several lakes respond differently in gradients of pH and light. We measured the growth rate of 12 genetically different strains from 5 lakes at 5 distinct pHs and varying light levels. The observed ability to grow over a wide range of pH and light levels helps to explain the successful spreading of G. semen. The additional strain specific preferences suggest adaptation to local environmental conditions, which might yield a competitive advantage over later invaders. Furthermore, these phenotypic differences might be an important driver for the genetic divergence into distinct populations.

THE ROLE OF PHENOTYPIC PLASTICITY IN FAST ADAPTIVE RESPONSES UNDER NATURAL CONDITIONS IN LAKE VICTORIA CICHLIDS

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Fast morphological changes in response to a changing environment are a common phenomenon in species that show adaptive radiation (e.g. Darwin finches, Anolis lizards). Most of these changes are attributed to directional selection rather than phenotypic plasticity. Lately, phenotypic plasticity and its role in adaptive radiation has attracted the attention of evolutionary biologists. The dramatic ecological changes in Lake Victoria during the past three decades provide a unique opportunity to study environmental effects on cichlid morphology. The environmental stressors are known to induce plastic responses in the lab but the mechanism behind adaptive responses under natural conditions remains obscure. The ecological changes in the lake include the upsurge of the predatory Nile perch, an increase of water turbidity, a decrease of dissolved oxygen (DO) levels and a diet shift of zooplanktivorous cichlids towards larger and more robust prey. We used ecological and morphological data collected in the past 30 years (at three year time intervals) to see how the environment has affected the morphology of the haplochromine cichlids. We found fast adaptive responses in four different haplochromines. In response to predation we found that the head-tail ratio decreased, probably to increase burst swimming speed. Unexpectedly, the eye size and double cone density decreased but the cone size remained unchanged. In response to the low DO levels, the gill surface increased. The cheek depth and premaxilla changed in a way that facilitates the manipulation of larger and more robust prey. In case of the latter, the changes found are of such proportion that they even cross subgenus boundaries. The found morphological changes reveal high similarity with changes found in plasticity experiments. We discuss the probability of phenotypic plasticity contributing to the morphological changes under natural conditions and the implications for the role of phenotypic plasticity in speciation.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

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INTRODUCING DIVERSITY? VARIABLE STRATEGIES IN NEW ENVIRONMENTS AND CONSEQUENCES FOR INFECTION DYNAMICS

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Invasion of new habitats is an important natural process known to require phenotypic plasticity of migrating individuals to cope with their new environments. It is poorly understood though how the potential for plasticity among individuals changes during an invasion. Increased trait variation among phenotypes of individuals experiencing a new environment could indicate the use of different strategies for coping with stress and a disruption of previously adaptive patterns. On the other hand, more uniform patterns in groups subjected to environmental changes would suggest a homogenizing effect of high stress levels. We investigate this question with a particular focus on immunity and immune related genes. The three-spined stickleback (*Gasterosteus aculeatus*) is known for its repeated and parallel invasion of different freshwater habitats from marine environments throughout the Northern hemisphere. In a lab-based study, we mimic transition from brackish water to freshwater and vice versa. Taking into account multiple biological levels as well as standing genetic variation and phenotypic plasticity, we explore the effects and patterns of variation caused by habitat invasion. Furthermore, we subject fish to experimental infection with the monogenean parasite *Gyrodactylus gasterosteus* and investigate differences in parasite susceptibility entailed by the mimicked invasion.

CONTRIBUTIONS OF A NOVEL POPULATION TO THE UNDERSTANDING OF THE ROLE OF MALE HYDROCARBON POLYMORPHISM IN DROSOPHILA MELANOGASTER

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In *Drosophila*, female hydrocarbons are known to be involved in premating isolation between different species and pheromonal races. However, the role of male-specific hydrocarbon polymorphism is not as well documented. The dominant cuticular hydrocarbon (CHC) in male *D. melanogaster* is usually 7-tricosene (7-T; C23:1), with the exception of central African populations, in which 7-pentacosene (7-P; C25:1) is dominant. Here, we describe a novel population from Comoro Island (Com) that includes males with sex pheromone profiles ranging from high 7-T to high 7-P. We maintained Com flies at different temperatures, without selection. After 18 months, flies reared at 21°C had a 7-T hydrocarbon profile and flies reared at 25°C had an intermediary to 7-P profile. We specifically selected for high 7-T, high 7-P, or intermediate hydrocarbon profiles. We showed that the 7-P/7-T ratio depended on temperature in Com with generally more 7-P at higher temperatures. There was partial reproductive isolation between flies with clear-cut phenotypes (7-T and 7-P). These results show that the dominant male pheromones are under environmental selection due to heat/desiccation stress.

GIFTS, COLORS AND THE VAGINA DENTATA: PHENOTYPIC PLASTICITY, HONEST SIGNALING AND SEXUAL CONFLICT IN A GIFT-GIVING BUTTERFLY

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Considerable recent effort has been devoted to understanding the roles of conflict and cooperation in sexual interactions. Gift-giving insects such as crickets and katydids have proven tractable systems for exploring these issues, with recent research emphasizing nuptial gifts as a source of conflict. In the Lepidoptera, males often transfer large nutrient-rich packages called spermatophores internally to females during mating. In contrast to work in other systems, researchers have typically characterized these nuptial gifts as cooperative contributions of mutual benefit to both partners. Male spermatophores provide essential nutrients that increase female lifespan and reproductive output. In turn, males benefit by delaying female remating and thus increasing their paternity share. However, males of many butterfly species, in an attempt to monopolize female reproductive output, package their spermatophores in tough chitinous envelopes. Females have, in response, evolved toothed structures in their reproductive tracts called signa, which serve to "chew" their way through the outer spermatophore coating to access the nutrients within. As a preliminary step in understanding the co-evolutionary dynamics in this system, we explored phenotypic variation in both male spermatophore quality and female signa morphology in the gift-giving butterfly Pieris rapae. We find that males honestly indicate the size and protein content of their spermatophores using sexually-selected wing color ornamentation. This relationship appears independent of larval access to dietary protein, suggesting that these two traits (coloration and spermatophore quality) abide by similar resource allocation rules. Female signa size and morphology was highly variable, scaled allometrically with body size, was independent of larval protein, and exhibited high narrow-sense heritability. We discuss these results in the context of sexual conflict and co-evolutionary dynamics.

THE NUTRITIONAL UNBALANCE ON FOREST PEST PERFORMANCE

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The spruce budworm is a major pest in the boreal forest of North America; this native insect exhibits eruptive behavior in approximately 30-year cycles. There is no explanation of what causes the populations in the forest to rise and fall; it could be due to coevolution processes. The continuous attack on host affects directly the nutritional balance of the buds, specially nitrogen and sugars. Under these conditions the insect's performance is affected so they are forced to go into an ecological evolution process. To avoid environmental variability, budworms were reared for tree generation on laboratory conditions and two artificial diets; a witness and an unbalance diet of N: C. We estimated heritability by parent-offspring regression analysis of the following traits: fecundity, developmental time, pupal mass weight, and the phenotypic frequencies of those traits. Results indicate that the population maintains substantial additive variance in life-history traits like other groups of Lepidoptera, and low heritability in pupal mass weight; more females presented higher percentage of fecundity on the unbalance diet in the last generation. This suggests that natural selection on spruce budworm is probably higher during the winter period so the high fecundities have to be maintained. This information is very useful in predicting the adaptation of this forest pest under epidemic conditions.

WATER FLEAS REQUIRE MICROBIOTA FOR SURVIVAL, GROWTH AND REPRODUCTION

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It is now generally accepted that microbiota play a major role in the proper functioning of their hosts. The use of model organisms and their bacteria has expedited the learning of these important roles by comparing conventionalized host-bacteria association with the bacteria-free host. In the model crustacean Daphnia, barely any knowledge exists as to the influence of microbiota on their physiology. We assessed if microbiota play a role in the fitness of D. magna by experimentally depriving them of their microbiota and compared their growth, survival and fecundity to that of their bacteria-bearing counterparts. We showed that bacteria-free hosts are smaller, less fecund, and have higher mortality than those with microbiota. We carried out these experiments on Daphnia coming from both a labreared parthenogenic clone and field-collected resting eggs (sexual eggs), demonstrating effects within and across host genotypes.

CORRELATED EVOLUTION OF SEXUAL DIMORPHISM AND MALE DIMORPHISM IN A LINEAGE OF NEOTROPICAL HARVESTMEN

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Secondary sexual traits (ornaments and weapons) increase male fitness, but are generally maladaptive when expressed in females, generating intralocus sexual conflict that is ameliorated through the evolution of sexual dimorphism. Intense sexual selection on males can also favor the evolution of male dimorphism, where alternative phenotypes that avoid expenditure in secondary sexual traits achieve copulations using 'alternative mating tactics' (AMTs). Secondary sexual traits can thus increase or decrease fitness in males, depending on which AMT they employ, generating a conflict within males that can be ameliorated by the evolution of male dimorphism. Thus, the phenotypic optima of females and small males are similar in terms of including the suppression of such secondary sexual traits. Male dimorphism could hence coevolve with sexual dimorphism, due to the evolutionary forces acting against both intralocus sexual conflict and conflict between males employing different AMTs. Here we tested this hypothesis by investigating the evolution of sexual and male dimorphism for two secondary sexual traits in 48 species of Neotropical harvestmen (Arachnida: Opiliones). Using a Bayesian approach with reversible-jump Markov chain Monte Carlo, we demonstrate that the two types of dimorphism present strongly correlated evolution, and that sexual dimorphism consistently precedes male dimorphism in this major arachnid group. Our findings were consistent for two different traits, and are robust to phylogenetic uncertainty. We propose that sexual dimorphism evolves earlier than male dimorphism because the genetic architecture for sex specific expression is already present even in sexually monomorphic species, due to sex chromosomes. The same is not true in the case for male dimorphism. We suggest that if a sexual trait arises first in an autosome and is expressed in all individuals, its suppression in females evolves more readily than its suppression in small males that adopt AMTs.

MODULARITY OF POLYPHENIC DEVELOPMENTAL REACTION NORM

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Whole boy of organism is the integrated entity, and it is an aggregation of multiple parts that have a certain level of independence and are functionally coordinated. The organismic integration recently becomes recognized as the heading of modularity. The integration moves ahead in the epigenetic developmental processes. Polyphenic developmental reaction norm makes distinct shapes among individuals, and it is a focus in evolutionary developmental biology and evolutionary ecology. It has been reported that larvae of salamander, Hynobius retardatus, exhibit a polyphenic developmental reaction norm within and among populations of high-low rearing densities. Above all, in high density, large-jaw cannibal-morphs are induced and they prey on the rest of the non-cannibal morphs. I investigated development of shapes, and the property of modularity in the shapes of the polypehnic reaction norm. I analyzed the shapes of hatchings (innate-morphs), low-density reared larvae (solitarymorphs), and high-density reared larvae (cannibal- and non-cannibal-morphs) according to the theory of landmark based Geometric Morphometrics. The Relative Warp Analysis and the thin-plate-spline shape presentations uncovered shape characteristics of each morphotype, some of which imply adaptive function. I found that all the samples coming form any of the ecological categories, i.e., innate-, solitary, cannibal, and non-cannibal, were completely discriminated on the shape-space that is defined by the Canonical Variates Analysis. Partial Least Squares Analysis indicated that the forward and the backward parts of body were separate modules in the innate-morphs and the solitary-morphs, on the other hand, the tow modules were integrated in the cannibal and the non-cannibal-morphs. This implies that a facultative modularity controls the polyphonic reaction norm.

THE ROLE OF OLFACTORY CUES FOR THE SEARCH BEHAVIOR OF A SPECIALIST AND GENERALIST BUTTERFLY

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Searching for resources is often a challenging task, especially for small organisms such as insects. To facilitate plastic and flexible responses complex stimuli have to be extracted from the environment, and the way different sensory modalities are integrated and translated into a relevant behavioral output is largely unknown. In order to entangle this processing, a first step is to investigate the relative roles of the different senses during search for various resources. While the role of olfaction is well documented in nocturnal moths, the olfactory abilities of the closely related diurnal, butterflies are poorly explored, even though physiological studies have revealed a well-developed olfactory system. Hence, in this study we tested two nymphalid butterfly species with divergent host plant range in a two-choice olfactometer. Results show that both the monophagous Aglais urticae and the polyphagous Polygonia *c-album* could navigate towards an invisible odor source, but behavioral variation was plastic and depended on task and intrinsic state. Unmated females of both species did not follow host plant cues, but mating shifted the behavioral response towards a preference for host plants. Floral scents could only be tracked by the nectar feeder *A. urticae*. Furthermore, the generalist *P. c-album* seemed to pay a cost for its higher plasticity in host use, by not to being able to make fine-tuned decisions between different combinations of host plants. We conclude that olfactory cues are important for the tested butterflies to navigate towards targets. We discuss possible limitations in processing capabilities of sensory input, underlining the need of synergistic effects between sensory modalities to facilitate and sharpen the search process.

PLASTICITY OF ANTIOXIDANT DEFENCE RESPONSES TO ABIOTIC STRESS IN NATURAL POPULATIONS OF IRIS PUMILA

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For plants, sunlight is not only the major source of energy for photosynthesis, but also a powerful evolutionary force that shapes the evolutionary trajectories of many traits. The aim of this study was to examine seasonal- and habitat-dependent variations in the specific activities of different antioxidants in the leaves of *Iris pumila* clones expressed under contrasting light conditions in the wild. Two populations were selected in the Deliblato Sands: one experiencing full sunlight and one from a woodland understory. Leaf samples were collected in spring, summer and autumn from the same clones in each population. The specific activities of the antioxidative enzymes, SOD, APX, CAT, GR and POD, and the contents of non-enzymatic antioxidants, anthocyanins and phenolics, were determined. All of the examined antioxidants were observed to be upregulated in summer-harvested leaves when compared to their spring or autumn counterparts. This suggests that reinforcement of the antioxidants might be the key mechanism for acclimatization of I. pumila leaves to seasonal variations in light intensity. In both populations, the antioxidative enzymes SOD, GR and POD expressed a greater mean seasonal plasticity than the other antioxidants. However, the between-population difference was significant only for POD. To elucidate whether variations in antioxidants reflect adaptations to local environments, a reciprocal transplant experiment was conducted in the wild. The higher activities of antioxidative enzymes were accompanied by increased contents of non-enzymatic antioxidants in both populations in the open habitat in comparison to clones in the shaded habitat. This could be a reflection of adaptive plasticity to the synergistic effect of strong light and high temperature. In contrast to seasonal plasticity, habitat-dependent plasticity peaked for APX and CAT in both populations. Apart from the phenolic content, a significant between-population difference was not observed.

WHEN DOES ENVIRONMENTAL CHANGE LEAD TO SELECTION ON PHENOTYPIC PLASTICITY?

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Continuous environmental change can lead to selection on the reaction norm slope if it alters the relationship between the optimal phenotype and environmental cues that determine the actual phenotype. For example, breeding time in small insectivorous birds, as e.g. great tits or flycatchers, is determined by temperatures in early spring while their reproductive success depends on insect phenology determined by temperatures in late spring. Differential changes of these two temperatures could alter the relationship between the cues used by the birds and the insect phenology and lead to selection on the slope of the birds' breeding time reaction norm. There are however a number of factors that could additionally affect the strength of this selection: It has been argued that in long-lived organisms plasticity would be more adaptive than in short-lived organisms because long-lived organisms experience a larger range of environments. Another relevant factor is temporal autocorrelation of environments: positive or negative autocorrelation will lead to environments experienced by an individual being more or less similar, which would affect the strength of selection on plasticity. The correlation between cues and optimal phenotypes also affects the strength of selection on plasticity as a low 'cue reliability' would select for less plasticity. Using simulation models we explored these effects in a scenario of a directionally changing environment focusing on phenological traits under climate change. Phenological traits are heritable but most genetic variation is present in the intercept of the reaction norm rather than the slope. Since the evolutionary potential of plasticity hence seems to be limited, strong selection on it would have negative demographic consequences. Consequently, it is important to know whether and how strongly environmental change would lead to selection on plasticity to better assess potential extinction risk caused by environmental change.

LIFETIME PATTERNS OF COOPERATION AND COMPETITION: BEHAVIOURAL SYNDROMES THROUGH TIME AND ACROSS CONTEXTS

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Individual differences in cooperative investment can be consistent and persist after life-history variation has been controlled for. This suggests that plasticity of cooperative behaviours may be limited, and individuals may be constrained to different behavioural trajectories or behavioural roles within animal societies. We use long-term observations of cooperative offspring care and mateguarding behaviours in a wild population of banded mongooses (Mungos mungo) to investigate lifetime patterns of behavioural consistency in a social context. Moreover, we examine patterns of consistent individual differences through time and across contexts to investigate what selection pressures may be driving consistent individual differences in this system. We find evidence for consistent individual differences in both cooperative and competitive behaviours, though the patterns of consistency are different for each behaviour. We suggest that changes in the costs of offspring care and the availability of mates that occur through time may drive variation in behavioural consistency seen between different age ranks. Individual differences in two forms of pup care are correlated, suggesting that individuals are not specialised to different cooperative activities, rather they may be specialised as helpful and selfish individuals. We find no evidence of correlation between individual differences in investment in offspring care and mate-guarding behaviours, suggesting that individuals do not show life time specialisations to roles as carers and breeders. This is one of the first studies to test lifetime consistency of behaviours in the wild, and may be the first to investigate consistencies in cooperative and competitive behaviours concurrently. Evidence for lifetime consistency of individual differences is suggestive of lifetime behavioural trajectories and advocates further study into early-life effects to determine the factors that lead to different individual trajectories.

DECIPHERING THE IMPORTANCE OF GENETICS AND ENVIRONMENT IN SHAPING THE PHENOTYPE OF LOW AND HIGH ALTITUDE VOLE POPULATIONS: COUNTERGRADIENT EFFECT AND CRYPTIC EVOLUTION

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Phenotypic variance along altitudinal gradients is determined by the influenced of both genetic and environment, and counter-gradient selection occurs when genetic and environment influences on phenotype oppose one another. Although we have previously found that high-altitude common voles (Microtus arvalis) are growing at slower rates and are lighter than low-altitude voles under standardized 'common garden' conditions (25°C), field based observations report no difference in body mass between high- and low-altitude populations. The heat dissipation theory has recently pointed out that cold ambient temperatures allow endotherms to increase their metabolic ceiling, and in turn favour for instance female lactation and offspring growth rates. Hence, one hypothesis is that genetically based differences in growth and body mass at adulthood between high- and low-altitude vole populations are masked in the wild by the cooler environments encounter by high-altitude voles, which might increase their metabolic ceiling and growth rates. To address this issue, Wild derived offspring (F1) of low and high altitude parents (F0) were reproduced at 10°C and 25°C in order to examine the influence of genetic (high vs. low altitude) and temperature on the growth of their pups (F2). We found that voles from high-altitude populations were growing at slower rates than those from low-altitude populations, but cool ambient temperatures allowed high-altitude voles to catch-up in size with lowaltitude voles. Our study highlights the occurrence of coutergradient effect, and in turn cryptic evolution, on the phenotype of an endotherm in response to temperature change.

METABOLISM AND ENERGY STORAGE STRATEGIES DIFFER IN NINE-SPINED STICKLEBACK (*PUNGITIUS PUNGITIUS*) POPULATIONS FROM CONTRASTING THERMAL ENVIRONMENTS

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Metabolic thermal adaptation is crucial for maintaining the necessary energy balance for survival in the face of seasonal temperature fluctuations. The plasticity of the thermal response and its potential for evolutionary adaptation are expected to differ between populations exposed to different thermal regimes. To understand how metabolic thermal responses differ in recently (< 11,000 years) isolated populations, we studied 4 populations of the nine-spined stickleback (Pungitius pungitius) from marine locations and spring-fed ponds from the Baltic region. We hypothesized that populations from cold, thermally stable ponds and from marine, thermally more variable locations would differ in their metabolic compensation thermal response. We acclimated wild-caught fish to 6, 11 and 19°C in the lab under constant photoperiod and measured their resting and active metabolic rates in order to determine their aerobic scope. Liver weight was used as a proxy for energy reserves. The fish from the coldest and most stable pond population exhibited complete temperature compensation for their aerobic scope while the marine populations underwent metabolic depression in the cold. Aerobic scopes were identical between all populations at 19°C. Marine populations had larger hepatosomatic index at all temperatures, with cold acclimation accentuating this effect. Our results suggest that differential energy storage and metabolism adaptations occurred in those populations, with the marine populations developing a winter-tolerance strategy while the cold pond population could potentially have a winterexploitation strategy. This work demonstrates the metabolic versatility of the nine-spined stickleback and provide a potential energetic framework for the previously observed adaptations in energydependent traits in other nine-spined stickleback populations.

DIFFERENTIAL GENE EXPRESSION IN THE OLFACTORY EPITHELIUM OF THE EUROPEAN EEL ANGUILLA ANGUILLA AT DIFFERENT LIFE STAGES

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The European eel *Anguilla anguilla* has a complex lifecycle that includes both freshwater and seawater stages and transoceanic migrations. Its ancestral line is believed to have split from the rest of the teleost lineage 225-325 million years ago. Although behavioral investigations have shown that olfaction plays a vital role in eel biology, little is known about the role of olfaction in the transition from fresh to sea water. Furthermore, the genes involved in eel olfaction have not yet been characterized. The first goal of this survey was to sequence and assemble the transcriptome of the olfactory epithelium (OE) of A. anguilla. Using 406 million paired-end reads, we assembled 417 669 contigs (306 Mb in length, mean contig length=732, N50=1281). A Blast search revealed that 89 631 of the assembly contigs have hits in Swiss-Prot. Among these are orthologs of type 1 and 2 vomeronasal receptors (V1Rs and V2Rs), trace amine-associated receptors (TAARs) and odorant receptors (ORs). The second goal was to look for differential gene expression in freshwater, seawater and sexually mature males. Our results show that eels make use of specific sets of genes at each of these three stages. Furthermore, the most drastic changes in gene expression occur in the OE of sexually mature males and include a large number of genes that are expressed at lower levels in this group. Though many genes are down-regulated in sexually mature males, we identified a number of receptors (e.g. V2Rs, TAARs and ORs) that are expressed at higher levels in sexually mature males and therefore maybe involved in reproduction (e.g. pheromonal communication). The results of this study will provide a thorough characterization of the genes involved in eel olfaction and will help to understand how changes in media influence changes at the molecular level in the OE. This research was supported by the Portuguese Fundação para a Ciência e a Tecnologia (PTDC/MAR/113608/2009).

VARIATION OF DROSOPHILA SUBOBSCURA WING MORPHOMETRIC TRAITS INDUCED BY 25 MT ELECTROMAGNETIC FIELD

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Human activities lead to extensive changes in ecosystems, with both ecological and evolutionary consequences. Electromagnetic fields of different frequencies are one of the most common and rapidly increasing environmental factors, but it is still unclear how organisms respond and/or adapt to such environmental changes. In an attempt to determine influence of every-day commonly used electromagnetic field (25 µT, 50 Hz) on morphometric traits, we analyze the direction and range of changes in wing size and shape in *Drosophila subobscura*. Wing development in *Drosophila* is well understood, and the wing vein network is excellent model system to investigate the effects of stressful environmental factors. The analysis was conducted using eight highly inbreed lines where one set of lines were reared in electromagnetic field, while another set of lines were reared in control conditions, over the three generations. We used method of landmark-based geometric morphometrics, which allows separating the shape and size effects, traits with different genetic properties and different response to the same environmental factors. Our data suggest that presence of extra low frequency electromagnetic field over extended period of time may reduce the stability of wing morphology and consequently reduce the fitness of exposed individuals. Therefore, specific stress that persists over multiple generations could increase the probabilities for extinction of populations composed of sensitive individuals.

ADAPTIVE PHENOTYPIC PLASTICITY IN AN ECOLOGICALLY RELEVANT FORAGING TRAIT

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The spectacular species richness of cichlid fishes and their famous diversity in morphology, coloration, and behaviour have made them a well-known textbook model for the study of speciation and adaptive evolution. In their natural environment, hypertrophic lip cichlids forage predominantly in rocky crevices. It has been hypothesized that this foraging behaviour associated with mechanical stress caused by friction could result in larger lips through phenotypic plasticity. In order to test how strongly phenotypic plasticity can influence the size and development of lips, we conducted a split design experiment in Nicaraguan cichlids and a series of breeding experiments on both Nicaraguan and African cichlids. Two months old full-sibs of A. labiatus (thick-lipped) and A. citrinellus (thin-lipped) were randomly assigned into two feeding groups, a control group (C) where food was released into the water column and a treatment group (T) fed with the same amount and type of food, but fixed to substrates in order to induce mechanical stress on lips. Treatment fish in the thick-lipped species had highly significant larger lips. Interestingly, no differentiation was found between treatment and control groups for the thin-lipped species. The thick-lipped species developed hypertrophic lips in both groups and these were significantly larger in both groups when compared to the thin-lipped species demonstrating a genetic component. The genetic component was further investigated by analyzing the phenotypic segregation in F1 and F2 fish obtained from a cross of thick- and thin-lipped species. F1 of both African and Nicaraguan crosses had intermediate lips and F2s exhibited large phenotypic variance, consistent with a polygenic basis. These results show that not only a genetic, but also a plastic component is involved in the development of hypertrophic lips in cichlids and opens the exciting possibility that plasticity is selected for in recent thick-lipped species.

PHENOTYPIC CONVERGENCE IN SEA BASS (*DICENTRARCHUS LABRAX*) ESCAPING FROM FISH FARMS: THE EFFECTS OF FERALIZATION?

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The impact of fish escaping from fish farms may depend on the extent to which escapees adapt to the natural environment, resemble wild conspecifics and become feral. Yet, little is known about the process of feralization in marine fish. We examined phenotypic changes in body shape, body condition and scale growth profiles of sea bass escaping from fish farms in the Canary Islands (where the species is rare or absent) and quantified the extent to which escapees had diverged from farmed conspecifics. Most feral sea bass had sizes that overlapped with those of farmed fish, indicating that they had escaped throughout the production cycle. However, 29% of escapees were larger than the maximum harvest size, suggesting that they had been at liberty for some time and had grown in the wild. Analysis of scale growth profiles showed that escapees grew at more variable rates than farmed fish in cages, but that some feral seabass were capable of growing in the wild as fast as farmed fish did. Feral sea bass tended to converge towards a similar body shape, having more streamlined bodies, lower body condition, and lower hepatosomatic indices than fish in cages. Although our study cannot discriminate between phenotypic plasticity and differential mortality of escapees, we interpret phenotypic convergence in feral sea bass as the likely result of a period of initial starvation, phenotypic plasticity, and selection against maladapted phenotypes. Our results suggest that plasticity may aid in the feralization process by allowing escapees to adapt quickly to natural conditions, and warn against the risks of rearing sea bass in open-net cages, particularly in areas where the species is not naturally found.

THE USE OF INDIRECT CUES IN ADAPTIVE COLOUR POLYPHENISM IN MOTH LARVAE

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Cues used to elicit adaptive plastic responses may be classified as direct and indirect. When a direct cue is used, phenotypes are adjusted in response to a factor which is an element of the selective environment. In the case of an indirect cue, the link between the cue and selective factor is less straightforward: the cue is used just to predict the occurrence of the actual selective factor. While direct responses can well be adaptive in novel environments, effective use of an indirect cue should imply that the cue and the selective factor must have co-occurred frequently enough in the evolutionary past of the species. We have shown that plastic responses in body colouration and patterning are based on combined use of direct and indirect cues in the larvae of a geometrid moth, *Ematurga atomaria*. The indirect character of some of the responses was revealed in an experiment in which larval colouration was shown to be adjusted to the appearance of the host plant also in absolute darkness; the cues are likely to be tactile. Such a response was unexpected because *E. atomaria* is a generalist herbivore not likely to have evolved specific responses to particular host plant species. The larvae of *E. atomaria* originating from geographic populations using different host plants showed analogous plastic responses which indicates that the link between the indirect cue and visual appearance of the host is, indeed, not limited to a particular host plant species. This case shows that the use of indirect cues for adaptive plastic responses needs not to be specific to a particular ecological interaction: *E. atomaria* appears to have evolved to associate rough surface of the host plant with 'patterned' environment at a quite general level.

NINE BACTERIAL SPECIES CAN'T BE WRONG: FLUCTUATING TEMPERATURE LEADS TO ASYMMETRIC CHANGES IN THERMAL TOLERANCE

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Fluctuating temperature is predicted to select for generalist genotypes that are capable of performing well across a wide range of temperatures. Although theories particularly predict fast fluctuations in selecting for thermal generalists, such experiments are scarce. Our aim was to find out whether fluctuating temperature selects for temperature generalists and test how uniform the temperature induced evolutionary changes are across different bacterial species. We set up a factorial experiment where ten replicate populations of nine different bacterial species were propagated separately either in a constant temperature (30 °C) or in a rapidly fluctuating temperature (2 h 20 °C - 2 h 30 °C - 2 h 40 °C, mean 30 °C). After 2.5 months we isolated altogether 720 bacterial clones from experimental populations and measured growth rate and yield (growth efficiency) in three constant temperatures (20, 30 and 40 °C). Meta-analysis of all of the species over all of the temperatures indicated that clones from the fluctuating temperature treatment had higher overall growth efficiency compared to clones from the constant environment. Moreover, the selection was found to be asymmetric, selecting more profoundly the tolerance of hottest temperatures. Generality of the results across studied species gives a strong support for the theories of evolution of thermal generalism but also indicates that evolutionary consequence of fluctuating temperature is especially strong in hot temperatures where the fitness consequences of increased or decreased heat are much more profound than in cold temperatures.

IS SOCIAL STATUS INFLUENCED BY INDIVIDUAL PROBLEM-SOLVING SUCCESS IN HOUSE SPARROWS (PASSER DOMESTICUS)?

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In group-living animals, group-members can benefit from having an innovative group-mate by scrounging or social learning. Therefore it may pay off for individuals to take group-mates' problemsolving abilities into account in social interactions such as aggressive competition and spatial association. We tested whether house sparrows' social status is related to their innovative skills in 15 small captive flocks. First we measured the birds' actual problem-solving success in food-extracting tasks. Then we manipulated their apparent problem-solving success while a new group-member (focal individual) had the opportunity to witness their performance. Later in group cages we observed aggressive interactions and spatial associations of the focal individuals with their group-mates. During the manipulation sessions, focal individuals paid more attention towards group-mates that spent more time on problem-solving attempts. Neither dominance rank nor spatial associations were related to either actual or manipulated problem-solving performance. However, focal individuals attacked more frequently those group-mates that had higher actual (non-manipulated) problem-solving success. This relationship depended on the focal bird's own performance, as less successful birds attacked their more successful group-mates consistently while more successful birds' aggression was context-dependent. These results suggest that house sparrows are able to assess their group-mates' problem-solving abilities; although they may use different phenotypic cues than those we manipulated. They may take this information into account in certain social situations, e.g. it may be advantageous for less successful individuals to scrounge aggressively from group-mates with better problem-solving skills.

HOW PLASTICITY IN SEXUAL ADVERTISEMENT AFFECTS SIGNAL RELIABILITY AND GENETIC VARIATION FOR SEXUALLY SELECTED TRAITS

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Exaggerated sexual traits can provide information to females about male performance, even if the precise alleles that confer high performance change along with environmental conditions. This plasticity in signalling may help to preserve genetic variation that would otherwise be eroded by strong mate choice, but it can also compromise signal reliability if environmental conditions change during development. We manipulated resource acquisition by altering the diet quality of inbred lines of decorated crickets (*Gryllodes sigillatus*) at both juvenile and adult stages. This allowed us to study both the effect of diet quality and a change in environment during development on trait expression. We measured a number of sexually and naturally selected traits in both sexes, revealing striking differences across diets in the expression of morphological and behavioural traits. We then assessed the reliability with which various traits signalled resource acquisition by assessing their genetic variance, as well as the covariance between traits and across environments. Our results show that traits such as body weight and calling effort, the male sexual advertisement trait, are more sensitive to environmental effects than morphological traits that are fixed at eclosion. We also show that there is positive genetic covariance between environments for calling effort, and high genetic correlations between the expression of calling effort in different environments. Substantial heritability exists for sexual advertisement such that it is a reliable indicator of the ability to acquire resources, for both stable and changing environmental conditions. The lack of evidence for genotype-by-environment interactions is inconsistent with a strong role for environmental diet quality in maintaining genetic variation required to sustain good-genes mate choice.

A COMMON GARDEN TEST OF LOCAL ADAPTATION IN BARNACLE POPULATIONS ALONG A SALINITY GRADIENT

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Environmental gradients may select for locally adapted populations. The barnacle Balanus (Amphibalanus) improvisus is found across a broad range of salinities and has during the past 150 years invaded the Baltic Sea with its gradient from fresh water to near marine salinity. This system may serve as a model of how marine organisms may respond to rapid environmental changes, e.g. climate change which is expected to reduce salinity in many coastal areas. We tested the hypothesis that local adaptations to low salinity have evolved during the past 150 years along the stable salinity gradient in the Baltic Sea. Common-garden experiments were performed to characterize physiological plasticity and possible local adaptations. Newly settled barnacles from each of three different geographical areas along the North Sea-Baltic Sea salinity gradient were exposed to different salinities (6 psu, 15 psu, 30 psu), and phenotypic traits including survival, growth, shell strength, condition index and reproductive maturity were recorded. The study showed that B. improvisus is a plastic and highly euryhaline species performing best at intermediate salinities, both in terms of maximum shell size and reproductive maturity. However, the results also indicate that low salinity has sub-lethal effects on fitness-related traits including slower initial growth and weaker shells. Overall, there were weak differences between populations in most measured traits, indicating little local adaptation to specific salinity conditions. However, some traits showed population-specific responses, e.g. that populations from high salinities obtained stronger shells in their native salinity compared to the other populations, possibly indicating local adaptation to different predation pressures. These results show that B. improvisus may be a rare example of a true brackish-water species, and that plastic responses are more likely than evolutionary tracking in coping with future changes in coastal salinity.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

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SEASONAL PHENOTYPE SPECIFIC GENE EXPRESSION DURING METAMORPHOSIS OF THE EUROPEAN MAP BUTTERFLY ARASCHNIA LEVANA

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The European Map Butterfly Araschnia levana represents the classic textbook example for seasonal polyphenism. This phenomenon occurs in species that respond to seasonally changing environmental parameters by formation of distinct phenotypes. A. levana occurs in two strikingly distinct seasonal phenotypes, the spring generation flying from April to June is reddish with black spots on the dorsal surface whereas that of the summer generation flying from July to August is dark-brown and labeled with a white band. Given that exposure of A. levana caterpillars to different light regimes decides whether metamorphosis results in pupae which are prepared for diapause and overwintering or not, we screened the transcriptomes of last instar larvae kept either under short or long day conditions for differentially expressed genes. Our results elucidate how environmental stimuli induce transcriptomic shifts which ultimately result in development of distinct phenotypes.

DEMOGRAPHIC AND BEHAVIOURAL LANDSCAPE OF DAMSELFISH REPRODUCTIVE SUCCESS

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Alternative reproductive strategies are common features in sexually reproducing organisms. For instance, in territorial species dominant males (having primary access to females) can be observed alongside other males adopting different strategies, such as parasitic fertilisation. In order for the alternative strategy to persist in a population, it has to hold some degree of reproductive success. In the case of the Mediterranean damselfish *(Chromis chromis)*, during the reproductive season, only territorial males colonise nest on the bottom of rocky reefs, and compete for females in order to receive egg deposition. Sneaker males float above the nesting area waiting for the right moment to parasitically spawn in the nests of territorial males. In this context, the success of sneakers is expected to be negatively affected by the ability of territorial males to recognise the rivals and chase them away. Variance in individual behavioural traits of nesting males as well as population-level factors, such as density can both affect the contribution of parasitic spawners to the next generation. Here we combine behavioural observation in the field with molecular analyses, using bi-parentally and maternally inherited markers to investigate the contribution of both individual chasing behaviour and population density on the relative reproductive success of nest-guarding males and sneakers.

QUANTITATIVE GENETICS BREEDING DESIGN OF THE WING SIZE AND SHAPE OF CACTOPHILIC DROSOPHILA SPECIES IN DIFFERENT ENVIRONMENT

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The interaction among insects and their host plants represent an important study model of genetic basics on phenotypic variation of quantitative traits regarding environmental change. This variation expressed by different environmental conditions can alter both the magnitude of environmental and genetics components of the phenotypical variation and the evolutive potential of a population, which is determined by the amount of genetic variability expressed by characteristics under selection. This study used a quantitative genetics breeding design in order to evaluate how a population of cactophilic Drosophila species (D. buzzatii cluster, repleta group) responds to a host cactus shift. This Drosophilacactus association is required during larvae feeding and development and some studies have shown plastic response in life history and morphological traits, considering different hosts. Data setting of 160 families (320 parents, 5760 offspring) from a full-sib analysis have estimated the heritability and genetic variance/covariance of wing size and shape regarding an ancestral and a new cactus. Results have shown the genetic correlation among traits was greater for the ancestral cactus; the additive genetic variance increased when larvae had been grown in a new environment, which could be explained by the expression of new genes that had not been subject to selection on the ancestral environment. Therefore, breeding Drosophila in a different host cactus imposes different selective pressures towards current genetic variation as well as relevant consequences for larvae development and adult morphology.

THE EFFECT OF RESOURCE AVAILABILITY ON HOST MANIPULATION IN THE COMPLEX LIFE CYCLE PARASITE SCHISTOCEPHALUS SOLIDUS – IS MANIPULATION COSTLY?

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Parasites with complex life cycles have to pass from one host to the next. They are often able to manipulate their host in a way that enhances transmission to the next host at the proper time point. It is largely unknown to what extent such manipulation might entail energetic costs for the parasite. If such costs exist, a parasite's ability to invest into manipulation might depend on resource availability. The tapeworm *Schistocephalus solidus* reduces the activity of its first intermediate host, a copepod, to lower predation risk before the parasite is ready to infect the next host, the three-spined stickleback. After becoming infective, S. solidus increases copepod activity to enhance predation and thereby facilitate transmission to sticklebacks. Using S. solidus and its copepod intermediate host, we investigated whether host manipulation is affected by the amount of resources available to the host. We subjected experimentally infected copepods to two feeding regimes and recorded their behavior (i.e. activity before and after a simulated predation attack). Additionally, we measured several unequivocal fitnessrelevant traits for individual parasites, including growth and development in copepods as well as infection success in and exploitation of sticklebacks. We will present our results on the influence of resource availability on host manipulation and how manipulation might affect parasite performance in its next host. This could give an indication of whether host manipulation by parasites is influenced by the host environment, potentially mediated by energetic costs of host manipulation.

HOW DOES DEVELOPMENTAL PLASTICITY CONTRIBUTE TO PHENOTYPIC EVOLUTION?

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The insect ovary consists of parallel repetitive units called the ovarioles, which are assembly lines for the production of eggs. The number of ovarioles is positively correlated with egg production rate and therefore is a morphological trait closely related to fitness. Interestingly, ovariole number exhibits both developmental plasticity and interspecific variation among Drosophila species. These two features of ovariole number provide an opportunity to investigate the contribution of developmental plasticity in evolutionary processes. To address this challenge, our approach is to compare the developmental mechanisms underlying the plastic response with those underlying genetic differences between related species. Here, we discuss how plasticity in ovariole number is regulated in Drosophila melanogaster. Since ovariole number is determined during larval development, we first described how nutrition affects this reproductive trait in carefully staged larvae. Our analysis revealed that early third instar larvae fed on sucrose alone show a stronger reduction in ovariole number than later stages, suggesting an early nutrition-dependent mechanism for ovariole formation. Further, we found that larvae fed on sucrose alone early in the third instar had ovaries with a reduced number of dividing somatic cells. Moreover, they show a delay in the onset of differentiation of the terminal filament cells, which serve as the starting point for ovariole formation. Currently, we are manipulating insulin and ecdysone signalling in the terminal filament precursors to determine the role of each in regulating the proliferation and differentiation of these cells to generate plasticity in ovariole number. This powerful approach will shape our understanding of how the environment creates phenotypic variation through changes in development and ultimately how environmentally-induced variation impacts evolutionary diversification.

BIODIVERSITY AFFECTS PHENOTYPIC PLASTICITY IN SUBTROPICAL TREES

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Plasticity allows plants to respond to environmental changes through changing phenotypes during their lifetime and this may be especially important for long lived species such as trees. Biodiversity loss could affect the expression of phenotypic plasticity of growth related traits and this could in turn affect the ability of species to respond to environmental changes. It is therefore important to understand what effects biodiversity has on phenotypic plasticity and we might expect two possible outcomes: firstly, more species rich communities might be more structurally diverse forcing plants to also express higher trait variation in diverse communities. Alternatively in low diversity communities niche space for individual species might be larger allowing niche expansion through an increased variation in growth traits between individuals. To test these ideas we studied plasticity of trees in a large biodiversity experiment in China "BEF China". In this experiment large scale experimental communities were established on an area of 24ha. A total of 60, 000 seed family replicates of thirteen subtropical tree species were grown in plots comprising 1, 2, 4, 8, 16 and 24 species. We measured total height, crown height, crown volume, stem diameter and herbivore damage in twelve species, across the diversity gradient. We calculated plasticity as the variance in growth traits between individuals of the same seed family within a plot. Our results show a significant and consistent decrease of growth trait variation with increasing species diversity. They indicate that biodiversity can influence the expression of phenotypic plasticity and they support the hypothesis that reduced interspecific competition causes increased trait variation. Trees in low diversity communities therefore seem to expand their niches by increasing trait variation.

INTRASPECIFIC AND INTERSEXUAL DIFFERENCES IN UV STRESS RESPONSE OF NEIGHBORING FRESHWATER COPEPOD POPULATIONS

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The role of phenotypic plasticity in local adaptation and thus speciation is controversial. On one hand, it may dampen the impact of disruptive or directional selection by producing adapted phenotypes without a heritable change in the genotype. On the other hand, it may facilitate population divergence because it enables an instant response to environmental change. Whereas theoretical studies have addressed this discrepancy, crucial aspects of the role of phenotypic plasticity during local adaptation have not been tested yet experimentally. We studied two neighboring populations of the copepod Acanthodiaptomus denticornis from the French lakes Lac Pavin (LP) and Lac de Montcineyre (LM), examining differences in carotenoid expression in response to UV stress. The populations were kept in climate chambers at standardized culture conditions. We found no genetic differences between the two populations at the mitochondrial COI locus. However, they differed significantly in their behavioral and physiological response to the stressor. LP males increased carotenoid expression from ~2 up to ~10 µg Astaxanthin per mg dry weight over a period of 14 days with regular exposure to UV radiation. LP females and LM specimens of both sexes, in contrast, maintained a concentration of ~0.4 µg Astaxanthin per mg dry weight under the same conditions. Moreover, the populations differed in their behavioral response to UV stress. While LM specimens showed no statistically significant movement in the water column after UV cue, LP specimens exhibit a pronounced downward movement. We hypothesize that lake-specific differences in water clarity and predation regime (presence of fish and insect larvae) in concert with sexual conflict drive the observed population divergence. We furthermore argue that the observed plasticity in carotenoid expression and the tightly coupled diverging antipredations strategies constitute a critical early step in an ongoing speciation process of *A. denticornis*.

VARIATION OF LIFE SPAN UNDER DIFFERENT ENVIRONMENTAL CONDITIONS IN DROSOPHILA MELANOGASTER COLLECTED ALONG AN ALTITUDINAL GRADIENT

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Geographical variations occur along latitude and altitude in diverse group of animals and plants. Latitudinal populations have received much attention, whereas altitudinal populations remain largely unexplored. We study with populations derived from 6 different altitude vary between 35 m and 2173 m with a short distance of 40 km between these two elevations point in Firtina Valley-Turkey. We test for a clinal trend in longevity under experimental conditions in order to test for geographical and/or environmental variation. Additionally we use nutrition and temperature stress for compare the degree of phenotypic variation relative to manipulated environmental conditions. Our results point out that, decrease of yeast concentration not extend life span in recently collected natural population from wild. However, as its known temperature is a important selective agent of longevity. Phenotypic variation varies between population for all experimental conditions and there was a significant interaction between temperature and population. Negative significant corellation occure between mean life span and altitude (r= - 0.143; p<0.0001) and nutrition (r= - 0.115; p<0.004) in extreme temperature (29°C), also in standart temperature the significant and negative corellation did not chang for nutrition (r= -0.117; p<0.004), but the significant correlation between altitude and life span changes (r = -0.071; p=0.055) in 25°C. Longevity seems to be population specific but not correlated with geographical origin in standard conditions. Populations under stressful temperature introduce a response correlated with geographical origin. Population longevity seems therefore to be independent from the experienced environments under standard conditions and indicating that this trait and its plasticity is population specific but not related to the geographical origin unless confronted with stressful condition.

TESTING EVOLUTIONARY PREDICTIONS OF THE PREDICTIVE ADAPTIVE RESPONSE HYPOTHESIS IN A PRE-INDUSTRIAL HUMAN POPULATION

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Environmental conditions during development can affect later-life health, but the evolutionary mechanisms underpinning these observations remain debated. The silver spoon (SS) hypothesis proposes that poor developmental conditions adversely affect development, leading to lower survival and fertility and increased metabolic disease risk, independent of later-life conditions. Meanwhile, the predictive adaptive response (PAR) hypothesis proposes that metabolic disease results from selection for development which is plastic with regard to environmental conditions, under which survival and fertility are maximised where conditions match in later life. If conditions change, metabolic disease results, but because the benefits of developmental plasticity exceed the costs, metabolic disease is not selected against. The observation that humans conceived during famine but raised in affluent conditions show later health problems is consistent with this idea. However, the proposed evolutionary mechanisms for putative developmental plasticity have rarely been empirically tested in humans. In particular, there are few studies examining the fitness consequences of conditions during development in varying later-life conditions. We tested the effects of early-life environmental conditions on later survival and fertility using data collected from several pre-industrial Finnish populations. We tested effects of early-life conditions on fitness during (i) varying later-life conditions, and (ii) adverse environmental conditions (a severe famine). Our results suggest that adverse early conditions decrease later fitness irrespective of later-life conditions, and that individuals born in poor conditions have lower survival and fertility during later-life famine. Our results do not support the PAR hypothesis, but are consistent with predictions of the SS hypothesis, which suggests that the early environmental conditions influence development in a manner which may persist into later life.

POPULATION DIFFERENTIATION IN PHENOLOGY OF THE ORANGE TIP BUTTERFLY IN SWEDEN AND THE UK

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Variation in environmental suitability for growth and reproduction throughout the year at temperate latitudes has given rise to intricate adaptations in the timing of life history events of flora and fauna in order to keep the life cycle in phase with the changing season. Phenological traits are often important for fitness, as they affect the biotic and abiotic environment an individual encounters and can vary geographically because of local adaptation as well as via phenotypic plasticity. Disentangling environmental effects (phenotypic plasticity) and genetic effects (local adaptation, genetic drift) behind phenotypic geographic variation of a species may aid in predicting ecological responses to changing environments. The spatiotemporal pattern of covariation of phenology and temperature in the orange tip, *Anthocharis cardamines*, was studied in order to partition the contribution of local adaptation and phenotypic plasticity to the specific traits. The butterfly shows geographic variation in its relation of phenology to temperature variation in the United Kingdom as well as in Sweden. This discrepancy in population differentiation may be driven by host plant phenology and availability, emphasizing the ecological importance of the temporal aspects of host plant-herbivore interactions.

THE ROLE OF DROSOPHILA INSULIN-LIKE PEPTIDES IN NUTRIENT-DEPENDENT LIFESPAN AND REPRODUCTION

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Dietary restriction (DR), a reduction in food intake without malnutrition, is a well-studied topic in ageing research. However, knowledge about mechanisms mediating this response is scarce. We know the Insulin-IGF Signalling (IIS) pathway is involved in ageing in many organisms. In Drosophila, insulin-like peptides (DILPs), upstream components of IIS, respond to different food conditions and a dilp2-3,5 knockout extends lifespan. This suggests that dilps can have a role in mediating the DR response. We investigated the role of dilps in nutrient-altered ageing by determining lifespan, fecundity and brain-dilp expression of the long-lived dilp2-3,5 knockout mutant (d235 Δ)and its genetic control (wDah) on a range of food types that differed in sugar and yeast concentration. We showed that $d235\Delta$ flies had an increased lifespan and decreased reproduction on all food types compared to wDah flies. There was still a nutrient-dependent lifespan and reproduction response for d235∆ flies, but the effect of the highest sugar and yeast concentrations was reduced for both lifespan and fecundity. For gene expression, in wDah flies dilp2 and -3 expression increased on high yeast and lower sugar concentrations, dilp5 expression increased on high yeast concentrations and dilp6 expression did not change on different sugar or yeast concentrations. Interestingly, in d235∆ flies dilp6 expression increased on high yeast concentrations. This data shows that dilp2, 3 and 5 may have a role but are not the key in mediating the DR response. We hypothesize that dilp6 expression may take over expression of dilp2, -3 and -5 in the d235 Δ flies, but not sufficient to maintain the same lifespan and reproduction on every food type.

FROM INDIVIDUALS TO POPULATIONS: LINKING THERMAL REACTION NORMS TO POPULATION DYNAMICS IN MICROCOSMS

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Long term studies of natural populations have shown that environmental changes can lead to shifts in life history traits (1) that can be difficult to predict due to complex ecological and evolutionary processes (2). Temperature is known to influence traits such as age or size at maturity, as represented by the temperature-size rule (3). The predictability of these modifications is still largely discussed (4,5). Indeed, individual level reaction norms can be overruled by ecological processes such as density dependence. Using the Collembola Folsomia candida, we described the reaction norms of maturation, growth and asymptotic size under a large range of temperature (6 to 26°C) and examined the structure and dynamics of populations reared under the same temperatures for three years. We used reaction norms to understand and predict variation of adult sizes and growth rates measured in the populations. The population dynamics and structure are shaped by temperature: a high temperature accelerates the life cycle and induces a change of the perceived food provisioning in the populations that alters the competition for the resource. But growth rate and body size in populations were found to be mainly determined by density rather than by the temperature itself. Under specific temperature, competition can trigger growth which leads to reduced population density. This underlines the need to untangle the complex interaction between environement and demography to help predicting how climate change impacts population dynamics. 1. A. Ozgul, S. Tuljapurkar, T. G. Benton, J. M. Pemberton, et al., Science 325, 464-7 (2009). 2. H. Kokko, A. Lopez-Sepulcre, Ecology Letters 10, 773-82 (2007). 3. M. J. Angilletta, Thermal adaptation: a theoretical and empirical synthesis (2009). 4. J. L. Gardner, A. Peters, M. R. Kearney, L. Joseph, R. Heinsohn, Trends Ecol Evol 26, 285-91 (2011). 5. J. A. Sheridan, D. Bickford, Nature Climate Change 1, 401-6 (2011).

LINKING PHENOTYPIC PLASTICITY IN DEVELOPMENT TIMES TO EVOLUTIONARY ORIGINS OF PROTANDRY: A META-ANALYSIS IN INSECTS

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Protandry is a widespread life-history phenomenon whereby males attain reproductive maturity before females. The explanations of the evolutionary origins of protandry fall into two broad classes. First, selection may favour earlier maturation of males per se, i.e. a sex difference in the timing of maturation may result from selective pressures on males and females to emerge at different times in relation to each other (direct selection hypothesis). Alternatively, the earlier maturation of males could be an incidental by-product of selection on some other sexually dimorphic trait, such as differential selection on body size of females and males (indirect selection hypothesis). In insects, protandry has been shown to be under direct selection in a few species, mostly in butterflies, while a more general understanding of the prevalence of direct vs. indirect selection on protandrous emergence remains unknown. One way to test between the two alternative hypotheses is to analyse the variation in the degree of protandry within species. It has been proposed that, for protandry to be directly selected, the degree of protandry should be insensitive to environmental conditions experienced during juvenile growth. This is because the optimal amount of protandry should be determined by adult life-histories, not by juvenile lifehistories. Here I will present the results of a meta-analysis on intraspecific, environmentally-induced variation in the degree of protandry with the aim to test between the direct and indirect selection scenarios of protandry. The analysis is based on a comprehensive literature-derived database on over 100 insect species, for which data on sex-specific development times and body sizes allowed to derive the response of protandry to environmental conditions.

CIRCADIAN FLUCTUATION OF GENE EXPRESSION ALONG A BATHYMETRICAL CLINE IN THE MARINE ANGIOSPERM POSIDONIA OCEANICA

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Plants develop mechanisms of adaptations at multiple levels to track and cope with fluctuations in the light environment. At molecular level, the correct matching of circadian variation of gene expression with environmental rhythms allows plants to optimize the utilization of environmental light and to prevent damages due to light excess. Several studies highlighted that a correct matching of endogenous rhythms and external rhythms increases organismal fitness over a board geographic range. Indeed, latitudinal clines in circadian clock gene expression levels (and polymorphism in clock genes) were found in plants, suggesting that natural variation in clock parameters are required to synchronize organisms with their specific environment. Understanding the genetic and physiological mechanisms that plants develop for the seasonal and daily response to environmental conditions, could allow to predict their response to unexpected changes in environmental conditions that could happen in the future due to anthropogenic and climatic changes. Here we explore variations existing, in the daily phase, along a bathymetrical cline in the marine angiosperm Posidonia oceanica (L.) Delile, a key species in costal Mediterranean ecosystems. To do that, we measured modulation of genes expression, by RT-qPCR, at six time points during the day, in several genes related to photosynthesis and circadian rhythms regulation in plants growing at three target depths during the daily cycle along a bathymetrical cline (5 to 30 meter depth). Sampling was performed in a continuous meadow located in the Bay of Calvi, Corsica (thanks to the ESF Cost Action 0906). We analyze the effects of the distinct environmental light conditions on the circadian fluctuation of gene expression. Further, we assess the phenotypic variation among and between genotypes and we discuss its potential adaptive relevance on P. oceanica fitness and survival.

INTERACTING PHENOTYPES AND THE MAINTENANCE OF BEHAVIOURAL PHENOTYPIC VARIATION IN FIELD CRICKETS

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Among the behaviours studied in the growing field of animal personality is aggression, and despite this trait being a product of an interaction between conspecifics, the effect of social partners on the focal individual has often been neglected. Since these ,interacting phenotypes' are simultaneously environments and evolving traits, social interactions may play a key role for maintenance of behavioural variation that is overlooked when considering the focal individual only. Here we investigate the extent to which the observed variance in behavioural phenotypes is due to social interactions using the field cricket gryllus campestri s. Male field crickets engage in agonistic contests with escalating stereotypical fighting sequences. We repeatedly measure aggressiveness of wild caught adult males in staged fights, where all individuals within a group encounter each other in dyads. We also take morphological measurements of traits known to affect contest behavior and outcome. We analyze aggressiveness in a behavioural reaction norm framework, which allows the integration of consistent differences in behaviours among individuals (personality) and their responsiveness to environmental variation (plasticity). By measuring behavioural and morphological traits in both the opponent and the focal individual, we also gain insight on the specific mechanisms influencing behaviours in a social context.

EVOLUTION OF PHENOTYPIC PLASTICITY IN CAENORHABDITIS ELEGANS POPULATIONS

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Theoretical models generally predict that phenotypic plasticity will evolve and be maintained if environments change across generations. *C. elegans* are known to tradeoff their ability to survive and reproduce under osmotic stress with progeny survival when the latter face anoxic stress. We used this paradigm to evaluate the importance of environmental stochasticity in the evolution of phenotypic plasticity. Specifically, populations adapted to osmotic stress were exposed to alternating or randomly fluctuating anoxic environments for 40 generations. Results for the experimental evolution of maternal effects on progeny survival under anoxia and normoxia will be presented.

SYMMETRIC AND ASYMMETRIC VARIATION ON DIFFERENT HIERARCHICAL LEVELS: PLASTICITY OF PETAL SHAPE IN HELIOSPERMA (CARYOPHYLLACEAE)

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Plants usually have a modular body plan, which consists of a number of building blocks (or modules). As sessile organisms, they are often confronted with spatially heterogeneous environmental conditions. Plastic responses may thus contribute to variation on several hierarchically organized levels: among parts within a module, among modules within an individual, among individuals within populations or species, and among populations or species. Flowers are particularly interesting, since most of them are composite modules consisting of several, repeated subunits, and within-flower variation can manifest itself as asymmetry. To assess the plastic component in flower shape variation, we examine whether directional environmental influences such as sun radiation and gravity produce a directional plastic response in morphometric traits. We compare several species of *Heliosperma* with 5-merous, radially symmetric flowers. Plants were grown in common garden, representative photographs of several flowers per individual were taken together with orientation and inclination data of each flower, and flat preparations of petals were made. Geometric morphometrics was used to characterize shape variation and asymmetry. Correlations of asymmetry with orientation and inclination of single flowers were evaluated and related to possible ecological functions and selective advantages. Furthermore, we quantified the amounts of variation associated with different levels – among and within species, among flowers within individuals, and between petals of one single flower. We discuss the role of plasticity for floral asymmetry, subindividual and higher-level variation.

SEXUAL SELECTION CAN IMPEDE THE EVOLUTION OF PHENOTYPIC PLASTICITY

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Phenotypic plasticity, the ability to adjust phenotype to the exposed environment, is an advantageous strategy for organisms to survive and reproduce. Although highly plastic organisms are rare, many previous studies reported small costs of phenotypic plasticity in various species. These studies argue for ecological costs, underlying direct genetic or physiological limits, or selection eliminating plasticity with high costs. Here we propose that sexual selection by assortative mating can impede the evolution of phenotypic plasticity in a plastic magic trait. We found that, when individuals plastically adapted to distinct habitats can mate and choose mates based on similarity in an ecologically diverging plastic trait (e.g., body size or foraging behavior), phenotypic plasticity can remain low even in the absence of physiological or genetic cost. This effect is likely to occur particularly at intermediate, empirically plausible strengths of assortative mating. This is because males with lower plasticity are more favored in mating between individuals adapted to distinct habitats, leading to stabilizing sexual selection that is sufficiently strong to counteract disruptive natural selection. Our finding suggests that research on the evolution of phenotypic plasticity should consider not only direct costs and limits but also how mating preference and phenotypic plasticity may mediate the directions and strengths of sexual and natural selection acting on the plastic trait.

5. Phenotypic Plasticity: Mechanisms, Ecology and Evolution

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THE FATE OF SEASONAL PLASTICITY UNDER RELAXED SELECTION

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Phenotypic plasticity of wing patterns is a widespread phenomenon among butterflies. Species occurring in seasonal habitats, where larval food plants dry out for an extended part of the year, often have a distinct dry season morph that is sedentary and delay reproduction until fresh plants are available. The dry season morph has a cryptic wing pattern to avoid detection by predators, while the wet season morph instead have conspicuous evespots to deflect predator attacks. The butterfly *Bicyclus* anynana is a model system for studies of phenotypic plasticity. In this species the plasticity affects more than just wing pattern so that the wet season morph allocates more of its available resources for quick reproduction. Wing pattern polyphenism is found in most *Bicyclus* species, but has not been well studied in species from less seasonal habitats. We studied the phenotypic responses to different rearing conditions in the rainforest species *B. martius* and compared them to *B. anayana*. The wing pattern responses were similar despite the fact that true dry season conditions never occur in the forest habitat. Other traits showed limited response, suggesting that *B. martius* is adapted for continuous breeding. Surprisingly, the relative reproductive investment from the larval resources was smaller than in wet season morphs of *B. anynana*. Knowing that *B. martius* is a long-lived butterfly this can be seen as an adaption for spreading investments over time while maintaining an optimal fat load for flight performance. Phenotypic responses are known to be under developmental hormonal control and it is thought that changes in hormone levels control a whole suite of correlated traits. In *B. martius* the life history traits seem uncoupled from the wing pattern polyphenism that still remains despite the presumed relaxed selection. This might be due to a relatively low fitness cost of expressing the wrong phenotype in a constant wet season environment.

ENVIRONMENTAL CUES AND GROWTH RESPONSES IN WILD YEAST

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Particular genotypes of wild yeast vary quantitatively in their ability to grow in particular environments in the laboratory. This basic form of phenotypic plasticity is modified by environmental cues, potentially interpreted in an 'anticipatory' way (e.g. an environmental factor activating a stress response that gives a phenotypic change resulting in better survival or growth under future stress). We have looked systematically for specific environmental cue – growth response relationships in sister species of wild yeast taken from the same environment (Saccharomyces cerevisiae YPS128 and S. paradoxus YPS138). In each case we constructed a cue-response network involving 98 stressors and metabolic substrates. Both species show intricate cue-response behaviour, differentiating between at least 50 environments. These networks have evolved substantially between the strains, far more than the evolution of their ability to grow in particular environments. The greatest changes involve galactose metabolism, where we identify transcriptional changes and a candidate genetic mechanism. Looking across the cue-response networks we find that broad network properties have also evolved. Contrary to a simplistic specialist/generalist distinction, the complete network in S. cerevisiae (a species known from many environments) is less complex than that in S. paradoxus (a woodland specialist). This raises questions about the evolutionary relationship between such behavioural plasticity and ecology, which we are now addressing in a wider variety of strains and species.

TEMPERATURE-PLASTICITY OF A SEED BEETLE - A SIZE-FECUNDITY RELATIONSHIP?

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The temperature-size rule states that organisms get smaller in warmer environments, and although the idea was originally developed for endotherms, the same pattern generally holds true also for ectotherms. Recent theory suggests that temperature-size rule for ectotherms may be explained by temperature-dependent size-fecundity relationships, so that individuals get larger in the temperature where the positive relationship between fecundity and size is steepest. We investigated this in 73 isofemale lines (originating from two African populations) of the seed beetle Callosobruchus maculatus, which is a capital breeder and adult size is dependent upon juvenile conditions. We subjected the larvae to two temperatures, 29°C (the temperature they are adapted to) and 36°C (close to the upper thermal limit). We found plasticity in size at emergence, such that individuals reared in 36° were larger than those reared at 29°C. Remarkably, the degree of plasticity was larger in individuals that were small at 29°C, indicating an optimum size at warm temperatures. In one population, we found that the positive relationship between size of males (the smaller sex) and was steeper at 36°C, with a similar trend in females, supporting the role of the size-fecundity relationship for the temperature size-rule in ectotherms. However, in the other population, no such relationship was found. This suggests that sizefecundity relationships could be driving the temperature-size rule in ectotherms, but that populationspecific factor, such as desiccation resistance, could also influence these patterns.

WILL KIDS BE WHAT GRANDPARENTS ATE? TRANSGENERATIONAL EFFECTS OF DIFFERENT FOOD QUALITY REVEALED BY DAPHNIA

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Daphnia are well known for their developmental plasticity, with phenotype modified as a consequence of parental environment, and this can be inherited through multiple generations despite clonal propagation. We examined whether multigenerational exposure of cladocerans to phosphorus (P) limitation alters the responses of the two next generations. Two clonal isolates, D.magna and D.pulex, were fed two standardized food quality (high and low carbon: phosphorus ratio) over 20 generations, resulting in four distinct lines. In reciprocal transplant experiment, we analysed a number of fitnessrelated traits for both daughters and grand-daughters of these lines. We focused on somatic growth rate, body RNA concentration (body growth rate marker), and the activity of alkaline phosphatase (phosphorus acquisition mechanism). As predicted by the Growth Rate Hypothesis, P-limitation had a strong negative effect on somatic growth rate, irrespectively of the species and line history. Here, we present evidence that food quality may have induced the transgenerational effects on somatic growth rate for one of the species used. The line origin for D.magna was highly significant for the diet response. Moreover, we found a significant interaction between diet type and line, which provides strong support for transgenerational effect. The responses were strong for D.magna but not for D.pulex, reflecting their different susceptibility to P-limitation. We also found a significant decrease in alkaline phosphatase activity related to multigenerational P-limitation stress. Currently RNA sequencing is performed to reveal the transcriptomic response underlying the observed responses.

Symposium

6. Evolutionary Consequences of an Early Germ-Soma Segregation

22 and 23 August



Program

Thursday 22 August Session(s): 9 Friday 23 August

Session(s): 10

Organisers: Duur K. Aanen and Rick Michod

Invited speakers: Norman Arnheim and Andrew Bourke

Description:

It has been 25 years since Leo Buss first proposed that an early segregation of a germ line from the rest of the body is an adaptation to limit the scope for selfish cell lineages. A number of recent discoveries make it timely to reevaluate this hypothesis. This symposium explores the evolutionary stability of multicellularity in organisms with and without an early germ-line sequestration, to discuss the relative importance of this characteristic for conflict resolution.

D22SY06IT10:30R5

MAJOR TRANSITIONS, THE EVOLUTION OF MULTICELLULARITY AND THE SIZE-COMPLEXITY HYPOTHESIS

Andrew Bourke¹

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The evolution of multicellularity is a prime example of a major transition leading to the evolution of individuality at a new hierarchical level [1-3]. As such, it exhibits many parallels with other major transitions, including the origin of eusociality in insects [4, 5]. Inclusive fitness theory represents a powerful tool for analysing the major transitions [4-6]. I discuss the evolution of multicellularity, including the evolution of a germline, in light of inclusive fitness theory. For example, the fact that most origins of multicellularity occurred via daughter cells remaining stuck to parent cells (subsocial route), and the likelihood that the first multicellular organisms had low cell numbers, suggest that cells within early multicellular organisms were clonal and exhibited few somatic mutations. This in turn suggests a near-identity of inclusive-fitness interests, implying that a germline would have evolved in such organisms not to prevent disruption from selfish cell lineages but to increase efficiency through a reproductive division of labour [4, 5]. The size-complexity hypothesis proposes that, as multicellular organisms grew larger, the increasing incidence of somatic mutations promoted the evolution of a segregated germline [5, 7]. The hypothesis predicts an association across taxa between a segregated germline and high cell number. I discuss evidence for this association, consequences of a segregated germline, and parallels with the evolution of eusocial insects.

- 1. Buss LW. 1987 The Evolution of Individuality. PUP.
- 2. Maynard Smith J, Szathmáry E. 1995 The Major Transitions in Evolution. Oxford: W.H. Freeman.
- 3. Michod RE. 2000 Darwinian Dynamics. PUP.
- 4. Queller DC. 2000 Phil Trans R Soc Lond B 355, 1647-1655.
- 5. Bourke AFG. 2011 Principles of Social Evolution. OUP.
- 6. Bourke AFG. 2011 Proc R Soc B 278, 3313-3320.
- 7. Reeve HK, Jeanne RL. 2003 Proc R Soc Lond B 270, 1041-1045.'ł

D23SY06IT10:30R5

WHAT IS ADVANTAGEOUS FOR THE GERMLINE MAY BE BAD FOR THE SOMA; THE IMPACT OF GERMLINE SELECTION ON THE MUTATIONAL LOAD IN HUMANS

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Some new germline mutations that arise in the testis may confer a selective advantage to the mutated germ cell relative to non-mutated cells. Theoretically, if a new mutation provided a germline selective advantage it could increase the frequency at which the mutated allele was introduced into the population by orders of magnitude even though, much to the species detriment, it reduced the fitness of the individuals that inherited it. We have shown examples of positive germline selection for three human disease mutations that arise sporadically each generation at frequencies ranging from 1/2,000 to 1/70,000 births. These sporadic disease cases occur at rates 100-1,000 times greater than would be expected based on what we know about genome average mutation rates. Using a testis dissection/mutation detection approach along with mathematical modeling we have shown that the high frequency of these *de novo* disease mutations cannot be explained by hyper-mutation at the diseasecausing sites. Instead, our data are consistent with the idea that the newly mutated germline stem cells have a proliferative advantage over non-mutated stem cells resulting in germline mosaicism. Plausible molecular mechanisms can explain the selective advantage for each of the three disease mutations. Others previously suggested that alleles conferring a selective advantage in the germline may be disadvantageous in the adult and might lead to "mitotic drive" systems that increase the mutational load of a population. The three disease mutations we examined may be realizations of this idea.

D22SY06RT11:18R5

GENOTYPES THAT CHEAT ON MULTICELLULARITY CAN EVOLVE QUICKLY IN FUNGI

<u>Eric Bastiaans</u>¹, Alfons JM Debets¹, Duur K Aanen¹ ¹Laboratory of genetics, Wageningen University, Netherlands eric.bastiaans@wur.nl

Multicellularity is a highly cooperative state prone to invasion by cheating genotypes that use the resources provided by the multicellular organism without contributing their fair share to nonreproductive functions of the organism (e.g. cancers). Kin selection, often realised through regular single -celled bottlenecks (and in some organisms by an early germline separation), is a solution to prevent selection for cheating. In fungi, the lack of an early germline separation and the potential to fuse with other individuals make cheating a realistic threat. However a genetic allorecognition mechanism that limits fusion to almost only clonally related individuals, seems to effectively protect fungi against cheating genotypes. In order to test the hypothesis that cheating is a realistic threat to multicellular growth in fungi, we used an experimental evolution approach with *Neurospora crassa*, that maximised the potential for cheating genotypes by selecting under low relatedness and completely local competition (i.e. under a high inoculation density of spores, in the absence of genetic allorecognition). Within less than 300 generations all eight replicate lines contained genotypes that matched our criteria for cheating: they had increased relative fitness (measured as proportion of spores produced) when in competition with a cooperative ancestral type, but spore production in monoculture was significantly decreased. So there is a clear trade-off between competitive fitness and production of asexual spores when grown alone. Contrary to predictions about the evolution of social behaviour that cheating genotypes will completely eradicate the social behaviour (the tragedy of the commons), we found a stable polymorphism in all evolved lines: a relatively cooperative type producing many spores when grown in monoculture, and the cheating type described above. We are currently studying the conditions leading to this apparently balanced polymorphism in our evolving lines.

6. Evolutionary Consequences of and Early Germ-Soma Segregation

D22SY06RT11:42R5

ALLORECOGNITION STABILIZES MULTICELLULARITY

Duur K Aanen¹, Tamas Czaran¹, Rolf F Hoekstra² ¹Laboratory of Genetics, Plant Sciences Group, Wageningen University, Netherlands ²Laboratory of Genetics, Wageningen University, Netherlands duur.aanen@wur.nl

The cells of a multicellular individual face the social dilemma of potentially increasing their personal fitness by increased reproduction at the cost of fitness of the multicellular individual. Organisms capable of somatic fusion are most sensitive to this somatic parasitism, since parasitic mutant cells can infect other individuals. Allorecognition, found in many multicellular organisms, limits the spread of somatic parasites. However, previous models have not satisfactorily demonstrated that this long-term benefit is sufficient to offset immediate disadvantages of reduced fusion experienced by new, initially rare, allorecognition types. Using a cellular automaton approach, we model the joint evolution of allorecognition and somatic parasitism in a multicellular organism resembling an asexual ascomycete fungus. Individuals can fuse with neighboring individuals, but only if they have the same allotype. Fusion with a parasite decreases the total reproductive output of the individual, but the parasite compensates for this individual fitness reduction by a disproportional share of the offspring. Our study shows that the mere threat of parasitism can select for high allorecognition diversity, which on its turn provides efficient protection against invasion of somatic parasites. Moderate population viscosity combined with weak global dispersal provided the best conditions for the joint evolution of allorecognition and stable multicellularity.

D23SY06RT11:18R5

CHANGE OF PARADIGM: AGING IS SELECTED FOR, ADAPTIVE AND PROGRAMMED

Kurt Heininger¹

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It is shown that the so-called "evolutionary theories of aging" are based on circular reasoning and that their basic assumptions are flawed. A non-group-selective, evolutionary mechanism is elaborated that explains the co-selection and programming of reproduction and aging/death [Heininger K (2012) The germ-soma conflict theory of aging and death: Obituary to the "evolutionary theories of aging". WebmedCentral AGING 3: WMC003275] Death of the soma is identified as the ultimate cost of reproduction. Importantly, germline cells control the longevity of the soma from 'within' by a variety of signals, e.g. sexual hormones. These signals limit the reproductive potential of the parent organism and drive a variety of aging pacemakers, particularly the senescene of the immune system. The transgenerational conflict between germline cells and soma over utilization of limited resources is the evolutionary rationale of postreproductive aging/death, semelparous organisms being a particularly drastic witness of the link between reproduction and death. Although the cost of reproduction, e.g. in terms of impaired immunocompetence and survival, still shapes the life history trade-offs of iteroparous organisms, the temporal uncoupling of reproduction and death concealed their evolutionary co-selection. In contrast to unitary organisms, modular organisms (e.g. plants, benthic aquatic invertebrates) that have no segregated germline and in which the adult body itself is a reproductive unit, may evade senescence. However, they are subject to territorial, density-dependent mortality patterns, due to e.g. self-thinning or chemical warfare, and density-limited seed recruitment driven by interindividual competition for resources. The germ-soma conflict shaped the different bauplans of unitary and modular organisms, is the motor driving animal coevolutionary Red Queen dynamics and fuelled the Cambrian explosion of animals.

6. Evolutionary Consequences of and Early Germ-Soma Segregation

D23SY06RT11:42R5

RELATEDNESS AND THE EVOLUTION OF MULTICELLULARITY

Roberta M Fisher¹

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It has been argued that a high genetic relatedness (r) between cells played a pivotal role in the evolutionary transition from single-celled to multicellular organisms. We tested this hypothesis with a comparative study, comparing the form of multicellularity in species where groups are clonal (r = 1), to species where groups are potentially non-clonal (r \leq 1). We found that: (1) only species with clonal group formation have undergone the major evolutionary transition to obligate multicellularity; (2) clonal organisms had more cell types, a higher likelihood of sterile cells and a trend towards higher numbers of cells in a group. More generally, our results build upon previous studies of animals, to show how group formation and genetic relatedness have played analogous roles in the three evolutionary transitions to multicellularity, cooperative breeding and eusociality.

POSTERS

D23SY06PS1266

CAN CHEATING STABILIZE ALLORECOGNITION? EXPERIMENTAL EVIDENCE IN FUNGI

<u>Eric Bastiaans</u>¹, Alfons JM Debets¹, Duur K Aanen¹ ¹Laboratory of genetics, Wageningen University, Netherlands eric.bastiaans@wur.nl

Multicellularity is a highly cooperative state prone to invasion by cheating genotypes that use the resources provided by the multicellular organism without contributing their fair share to non-reproductive functions of the organism (e.g. cancers). Kin selection, often realised through regular single-celled bottlenecks (and in some organisms by an early germline separation), is a solution to protect against cheating. In fungi, the lack of an early germline separation and the potential to fuse with other individuals make cheating a realistic threat. However a genetic allorecognition mechanism that limits fusion to almost only clonally related individuals, seems to effectively protect fungi against cheating genotypes. We have shown earlier that in the absence of cheating genotypes, the fitness advantage of fusion selects against allotype diversity in the fungus *Neurospora crassa*. Individuals with more common allotypes have a higher fitness because they fuse more frequently and gain a larger average size. Studies that model evolution of allorecognition in fungi show that cheating can cause a stable polymorphism for genetic allorecognition loci. Using cheating genotypes generated during an experimental evolution experiment, we empirically test the hypothesis that genetic allorecognition in fungi can be stabilized by the presence of cheating.

D23SY06PS1279

HAPLODIPLOIDY AND THE EVOLUTION OF EUSOCIALITY: WORKER REVOLUTION

Joao Alpedrinha¹, Andy Gardner¹, Stuart West¹ ¹Stuart West, Zoology, University of Oxford, Portugal joao.alpedrinha@gmail.com

Hamilton suggested that haplodiploidy predisposes species to the evolution of eusociality. This is because haplodiploidy increases the genetic similarity of sisters above that of mother and daughter, which could potentially favour an individual to give up her own future reproductive success in order to devote her life to sib-rearing. Trivers and Hare noted that, in order for this to work, helpers need to direct their altruism preferentially towards sisters rather than brothers. Building upon this idea, they proposed two biological scenarios whereby haplodiploidy could promote eusociality: (a) workers biasing the sex allocation of the queen's brood towards females; and (b) workers replacing the queen's sons with their own sons. However, biased sex allocation and worker reproductive values of males and females. Here we determine the net effect of all these consequences, for the two scenarios whereby the workers seize control of reproduction. We find that: (1) worker control of sex allocation may promote helping, but this effect is likely to be weak and short-lived; and (2) worker reproduction tends to inhibit rather than promote helping.

Symposium

7. The Nature and Mechanisms of Evolution of Species Recognition Systems

20 and 21 August



Program

Tuesday 20 August Session(s): 4 Wednesday 21 August Session(s): 5, 6

Organisers: Guila Ganem and John Endler

Invited speakers: Tamra Mendelson and John Endler

Description:

What makes a mate recognition system specific? Is species recognition a good working concept? How does specificity evolve? Do the mechanisms involved differ in the presence or absence of gene flow? These questions are central to our understanding of evolution of behavioral isolation, one of the most important means of species diversification, and the symposium goal is to address them.

D20SY07IT17:45R9

WHAT IS "SPECIES RECOGNITION"?

Tamra Mendelson¹

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The concept of "species recognition" is widely used in the study of animal communication, typically referring to the tendency of organisms to distinguish members of their own species from heterospecifics. Yet, it is difficult to pinpoint a consensus definition of the concept, and close inspection reveals a host of underlying assumptions. First, I discuss definitions and underlying assumptions of species recognition as used in the literature and identify where these assumptions are either untested or logically flawed. I then discuss the implications of species recognition for the question of whether non-human animals have species concepts and articulate directions for research into the cognitive architecture of species boundaries in nature.

D21SY07IT10:30R9

THE MULTIPLE FUNCTIONS OF SEXUAL DISPLAYS AND THEIR IMPLICATIONS FOR SPECIES RECOGNITION SYSTEMS

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Sexual displays have functions other than indicating potential mate quality. I will briefly discuss them and give an example in an Australian bowerbird. I will then discuss their implications for species recognition systems, speciation, and the generation of species diversity.

D20SY07RT18:33R9

INNATE SEX RECOGNITION AND LEARNED SPECIES RECOGNITION IN AN INVASIVE FISH

Kit Magellan¹

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Sex and species recognition are both fundamental to sexual reproduction. Speciation imposes modification of species recognition traits but not necessarily those for sex recognition. However, most studies either focus on sex recognition characteristics in a single species or assume sex recognition a priori and focus solely on species recognition traits. Invasive species represent an ideal opportunity to explore the relationship between these two recognition systems. I investigated interactions between the highly invasive mosquitofish, Gambusia holbrooki and the native Iberian toothcarp, Aphanius iberus. I found that male Gambusia consistently differentiated between male and female heterospecifics but initially failed to distinguish heterospecific females from those of their own species. However, they began to learn to recognise species differences within 24 hours. Given the lack of costly mating displays and nuptial gifts in the mosquitofish mating system, sex recognition and misdirected mating attempts may be less costly than species recognition. This and their rapid learning ability may be a factor explaining the invasive success of G. holbrooki.

D20SY07RT18:57R9

SPERM SWIMMING ABILITY AND REPRODUCTIVE ISOLATION IN FICEDULA FLYCATCHERS

<u>Murielle Ålund</u>¹, Simone Immler², Anna Qvarnström¹

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Incomplete premating isolation can lead to costly hybridization in sympatry. When premating barriers are absent or incomplete, postmating prezygotic isolation (or gametic isolation) can prevent heterospecific fertilization. For example, poor transfer and/or storage of sperm, low sperm competitive ability or incompatibility between gametes can contribute to low fertilization success between heterospecific individuals. Sperm traits (e.g. viability, longevity, swimming velocity and motility) play a crucial role for fertilization success and divergence in any sperm trait between species could facilitate prezygotic isolation. Here we compare sperm swimming ability, motility and longevity between two hybridizing species of flycatchers (*Ficedula albicollis* and *F. hypoleuca*) from a well-studied hybrid zone population on the island of Öland (Sweden), in the Baltic Sea. Species recognition is imperfect in this system and leads to costly hybridization. Indeed, about 4% hybrids with strongly reduced fertility are produced every year in our study population. We relate our measurements of ejaculate quality to the fertilization success of flycatcher males in different crosses within and between species. As density and competitive ability differ between these two flycatcher species, asymmetric gametic isolation is expected in the system. We discuss the implications of our results for the rate and direction of gene flow between pied and collared flycatchers.

D21SY07RT11:18R9

SPECIES RECOGNITION IN HETEROGENEOUS SENSORY ENVIRONMENTS

<u>Gwendolen M Rodgers</u>¹, Marc Théry¹, Mireille Belle Mbou², Florence Bayle¹, Stephane Sourice², Secondi Jean²

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Reproductive isolation is central to the speciation process and efficient species recognition is crucial to prevent the waste of time, energy and resources in low quality matings. Ideally, animal communication systems are tuned to environmental conditions, facilitating the selection of appropriate mates. However, breeding individuals often face heterogeneous conditions some of which may reduce preclude efficient species recognition and enhance the hybridization rate. Recent work suggested that information may be reliably transferred under a broad set of environmental conditions by resorting to multimodal communication (backup hypothesis). These predictions may not hold for species experiencing recent environmental changes but they may do for species experiencing long term stochasticity of their sensory environment. The two newts Lissotriton vulgaris and L. helveticus extensively overlap in Europe. Mating involves tactile, visual and chemical components. We investigated how the sensory environment affected species recognition and whether the use of multimodal sexual communication supported the backup hypothesis. We showed that despite large phenotypic differences, hybridization is common at the site level. We also found that visual and olfactory communication contributed to recognition under ideal transmission conditions but that visual recognition was drastically reduced under poor, but realistic, visual conditions. We thus tested whether olfactory recognition would maintain a high recognition level regardless of visual conditions. We also tested whether the visual environment experienced during development influences visual perception in order to determine the level of constraints for the olfactory channel. These results show that variable sensory environments may contribute to long-term hybridization. They also suggest that selection against hybrids may generate divergent habitat selection and allow overlaps only in habitats allowing efficient recognition.

D21SY07RT11:42R9

EVOLUTION OF ACOUSTIC AND VISUAL SIGNALS IN ASIAN BARBETS

<u>Alejandro Gonzalez-Voyer</u>¹, Alberto Castelló¹, Jennifer Leonard¹

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The study of animal communication systems is an important step towards gaining greater understanding of the processes influencing diversification because signals often play an important role in mate choice and can lead to reproductive isolation. Signal evolution can be influenced by a diversity of factors such as biophysical constraints on the emitter, the signalling environment, or selection to avoid heterospecific matings. Furthermore, because signals can be costly to produce, trade-offs may exist between different types of signals. Here, we apply phylogenetic comparative analyses to study the evolution of acoustic and visual signals in Asian barbets, a clade of non-Passerine, forest-dependent birds. Our results suggest that evolution of acoustic and visual signals in barbets is influenced by diverse factors, such as morphology and signalling environment, suggesting a potential effect of sensory drive. We found no trade-offs between visual and acoustic signals. Quite to the contrary, more colourful species sing significantly longer songs. Song characteristics presented distinct patterns of evolution. Song frequency diverged early on and the rate of evolution of this trait appears to be constrained by body size. On the other hand, characteristics associated with length of the song presented evidence for more recent divergence. Finally, our results indicate that there is a spatial component to the evolution of visual signals, and that visual signals are more divergent between closely related taxa than acoustic signals. Hence, visual signals in these species could play a role in speciation or reinforcement of reproductive isolation following secondary contacts.

D21SY07RT14:00R9

FROM SEXUAL COMMUNICATION TO SPECIES RECOGNITION: EXAMPLES FROM THE HOUSE MOUSE

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Sexual communication involves transfer of information between potential mates on their identity, quality, compatibility and history. Both endogenous and exogenous factors are expected to shape the evolution of the complex systems made of signals, receptors and preferences. Further, variation in the latter factors could result in divergence between the sexes, populations and species. How potential mates make sense of the diverse information available for mate choice remains an unresolved question. My research addresses this question in rodent species using the olfactory chemosensory channel to communicate. For example, several pheromones involved in social and sexual communication have been described in the house mouse (Mus musculus domesticus). These chemical cues are mostly present in the mouse urine and the mouse marks its territory by depositing urine drops. Moreover, mice are both excellent noses and carry odor signatures that act as fingerprints. Still, despite of this remarkable individual variability population and species differences exist. The house mouse is involved in chromosomal diversification in parts of its range and share a hybrid zone with another subspecies (M. m. musculus) along a north south axis crossing Europe from Scandinavia to the Black sea. Using these different evolutionary and geographical settings, research in my laboratory has addressed the mechanisms involved in mate recognition system divergence. Referring to examples from my research I shall illustrate how genetic drift, local adaptation sexual selection and reproductive interference shape the house mouse mate recognition system and could facilitate speciation.

D21SY07RT14:24R9

WHY IS THERE SO LITTLE PRE-COPULATORY CHOICE IN A PROMISCUOUS SEED BUG?

<u>Emily R Burdfield-Steel</u>¹, David M Shuker¹, Liam R Dougherty¹ ¹School of Biology, University of St Andrews, United Kingdom erb28@st-andrews.ac.uk

Male bugs of the family Lygaeidae have been recorded to sexually harass and even mate with females of different species, both in the wild and in the laboratory. Studies have shown that female bugs suffer fitness costs from repeated male harassment, and that this may even be the case when this harassment comes from heterospecifics. We present recent findings that suggest that these inter-specific mating attempts are not the result of an inability to distinguish between conspecifics and heterospecifics, but rather a failure to use that information to inform mating decisions. We suggest that the available data on the mating system of these seed bugs may explain this pattern. For instance, males appear able to correctly differentiate conspecifics from heterospecifics under some circumstances, and will do so during interactions that require greater investment (i.e. mate guarding). As most seed bug species live in patchy habitats, encounter rates in the wild may be low and selection may act to reduce male choosiness during the early stages of copulation in order to avoid missing out on potential mating opportunities. The lack of any obvious pre-copulatory courtship supports this model of opportunistic mating. Additionally, the evolution of pre-copulatory courtship and signals may be constrained due to the aposematic nature of many seed bug species. This may mean instead that post-copulatory sexual selection, including a high frequency of mating failures, may be key determinant of non-random mating in these species.

D21SY07RT14:48R9

DOES INCORPORATION OF ALARM CALLS INTO AVIAN SONG INCREASE RATES OF SIGNAL DIVERGENCE ACROSS SPECIES?

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Assortative mating of co-occurring species depends on the interaction between the evolution of signals involved in mating decisions and the corresponding recognition mechanisms. In two hybridizing *Ficedula* flycatcher species, both plumage characteristics and mate preferences are found on the paternally inherited Z chromosome, suggesting that assortative mating is achieved in these species through physical linkage of signals and preferences. In flycatchers, as in other songbird species, song functions not only in mate attraction, but also in interactions between males and has a learned component, leading to a series of potential problems for genetic linkage and assortative mating. Here, through experimental playbacks to *Ficedula* chicks from each species still in the nest, I show that chicks respond more strongly to the songs of their genetic fathers, suggesting that recognition of complex vocal signals is determined by genes on the Z chromosome. In contrast, chicks respond readily to the innately produced alarm calls of the other species, suggesting that recognition of vocal signals is achieved through mechanisms that are independent from how they are transmitted.

D21SY07RT15:12R9

ENTAMOEBA VARIETIES USE BIOCHEMICAL SIGNALING AND BEHAVIORAL AGGREGATION TO DISCRIMINATE BETWEEN MEMBERS OF DISTINCTIVE STRAINS

<u>Avelina Espinosa</u>¹, Guillermo Paz-y-Miño-C² ¹Biology, Roger Williams University, United States ²Biology, University of Massachusetts-Dartmouth, United States aespinosa@rwu.edu

Evolutionary processes in which selection acted continuously and cumulatively on ancestors of Entamoeba populations gave rise to chemical and behavioral signals that allowed individuals to discriminate non-population members and, gradually, to the emergence of new lineages. The concept of 'species recognition' at the unicellular level might be artificial and inadequate to define signaling in single-cell natural populations. Aggregative behavior could be explored in a nonsocial protist to define discrimination cues among/between natural varieties. We demonstrate that by color tagging and pairmix-culturing six *Entamoeba* varieties, the difficulty of discerning among apparently similar taxa can be resolved. When grown together with different amoeba strains, free-living/opportunistic (*E*. moshkovskii Laredo), commensal (E. moshkovskii Snake) or parasitic (E. invadens IP-1, E. invadens VK-1:NS, E. terrapinae, E. histolytica) trophozoites aggregate only with members of their own lineage. Clusters of trophozoites from each amoeba show distinctive rate of aggregation, density of cells per cluster, and distance between clusters. By using these behavioral cues, and identifying the genes involved in cell-signaling for cluster formation, distinctive amoeba taxa can be characterized quantitatively; we postulate that not only *Entamoeba* varieties, but apparent taxa crypticity in other protists, can be resolved by examining the natural ability of unicellular eukaryotes to discriminate between members and non-members of a lineage. Thus, phylogenetic relations among protists, which are usually determined by morphology and molecular techniques (the latter often confounded by horizontal gene transfer), could be further understood by incorporating behavior into the evolutionary analysis of this complex group of organisms.

POSTERS

D20SY07PS0337

PREMATING REPRODUCTIVE ISOLATION BETWEEN TWO CLOSELY RELATED CRICKET SPECIES

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Reproductive isolation (RI) is the key requirement for speciation and, as a possible first isolation barrier, premating RI can be particularly important during early speciation stages. In our study we address premating RI between Gryllus texensis and G. rubens, two closely related sympatric species of field crickets. Male crickets attract females with a calling song, a cue used for species recognition. Previous studies have shown that the two species differ in pulse rate of the song and in female preference for pulse rate, but not much is known about other song parameters or about hybrid preference. In this study we discuss the following two questions: (i) which (other) parameters of the calling song play a role in premating isolation; (ii) which part of the song (and song-preference) parameter space do laboratory-bred F1 and F2 hybrids occupy? For both species, male songs were individually recorded in an anechoic chamber to analyze the within and between species variation for several temporal parameters and for carrier frequency of the song. Female preference was measured using two set-ups. First, two-choice arena experiments were used to test the strength of the preference for either con- or heterospecific (artificial) songs broadcasted by two juxtaposed speakers. Second, by mounting a female upon a sphere opposite to two orthogonally placed speakers, we tested her preference for a range of stimuli. Pulse rate (pulses s-1) and duty cycle (duration relative to the period) were the most important temporal parameters for species separation. Additionally, preliminary results showed that *G. texensis* preferred a higher carrier frequency than *G. rubens*, while males of both species sang at a similar frequency. Hybrid song and preference had intermediate values, with preliminary results indicating a bias towards the maternal parent species.

D20SY07PS0349

RAPID DIVERGENCE IN MATING BEHAVIOR VERSUS SEXUAL MORPHOLOGY AMONG DIVERGING POPULATIONS IN A WIDESPREAD NEOTROPICAL FLY, ARCHISEPSIS DIVERSIFORMIS (DIPTERA: SEPSIDAE)

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Reproductive traits often evolve rapidly and can be important in generating sexual isolation, restricting gene flow between diverging populations. In particular, pre-mating discriminating mechanisms, such as courtship behavior, can evolve faster than other types of traits. Here, we present a detailed study of mating behavior and sexual morphology in two populations (Costa Rica & Panama) of the widespread neotropical sepsid fly *Archisepsis diversiformis*. We find that (i) despite overall similarities in courtship repertoires, some behavioral elements performed during mating are clearly population-specific, and (ii) these populations exhibit clear pre-mating isolation when tested one-on-one. Nevertheless, masscontainer population crosses (perhaps similar to situations that occur in the wild) did produce viable F1 offspring after extended exposure. (iii) Furthermore, morphometric analysis indicates that the populations differ significantly in male and female wing shape but only moderately in the shape of the grasping structure on the male fore femur, and not at all in the male genital clasper shape. (iv) Finally, a comparison of the fast-evolving cytochrome oxidase subunit I (COI) gene fragment indicates that individuals from Costa Rica & Panama are genetically highly similar, forming a strong monophyletic cluster with uncorrected pairwise distances ranging from 0.5-1.6%. This study documents that behavior diverges faster than morphology, which may play a role in impeding gene flow between these two populations. We argue that fine-scaled behavioral work is important when studying incipient sexual isolation and ongoing processes of speciation among widespread species.

D20SY07PS0783

ASYMMETRIC REPRODUCTIVE INTERFERENCE AMONG TWO SPIDER MITE SPECIES

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Incomplete specific recognition can lead to the occurrence of reproductive interference (RI) reproductive interactions between two species resulting in fitness loss for at least one of them. RI can play an important role in the coexistence of species, being especially important in the fate of introduced exotic species and in pest management. Tetranychus urticae and Tetranychus evansi are two closely related haplodiploid spider mite species that often coexist in solenaceous crops. Incomplete specific recognition occurs among these species: heterospecific matings were observed, although no hybrid progeny has been found. We tested two possible RI mechanisms: (1) the effect of mating with heterospecifics on virgin (haploid) offspring and (2), the consequences of heterospecific crosses for the offspring of females that have or will mate with conspecifics. Behavioural assays showed that (1) only T. urticae females and T. evansi males prefer to mate with conspecifics; (2) regarding latency to copulation individuals behave as virgins after mating heterospecifically, (3) *T. urticae* females copulate for a shorter period with heterospecifics than with conspecifcs. Results for fecundity and sex-ratio revealed that (1) for both species, fecundity of females mated with heterospecifics are similar to that of virgins - heterospecific crosses do not affect egg viability; (2) T. evansi females that mate with both conspecific and heterospecific males had higher fecundity than females that mated with a conspecific male only; this was not observed in T. urticae. (3) T. urticae females that mate with a heterospecific male after a conspecific mating had a lower percentage of female offspring. The results obtained point to the occurrence of asymmetric RI, in which *T. evansi* females benefits from mating with heterospecifics, whereas *T. urticae* pay a cost of such matings. These results may affect the coexistence of these species, a hypothesis requiring further testing.

D20SY07PS0874

HOW CAN A SEXUALLY SELECTED EXAGGERATED MALE TRAIT BE REDUCED OR LOST?

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What drives the evolution of male specific conspicuous traits? Sexual selection is known to act via female preference for exceptional traits. However, using phylogenomic (RAD sequencing) analysis we have shown that a definitive example of such a trait, the exaggerated caudal fin (or sword) specific to *Xiphophorus* fish, is likely to have been lost during the evolutionary history of this genus. If such a trait is subject to strong sexual selection, how can it be lost? To address these questions we used the green swordtail fish, *Xiphophorus hellerii*, whose sword is a textbook example of female association preferences. Classic studies have demonstrated through association tests that females prefer males with long swords, and even species whose males lack this trait prefer males with an artificial sword over their own non-sworded males. Here, we extend these findings by using a natural population setting, where males and females could freely interact and choose mates, to show that female preferences are highly likely to involve an array of male traits. By conducting independent replicate experiments we find no significant difference in paternity success between long and short sworded males, and in any given replicate females tended to choose the same male. Further, by genotyping sperm stored in the female ovary we provide the first indication that post-copulatory sexual selection may play a role in the paternity success of different males. Previous studies have largely painted a picture of the sword as the main determiner of reproductive success. Under this scenario, the repeated loss of the sword is paradoxical. By considering male sexiness as a more multivariate trait these losses can be explained by changes in the environment and fluctuations in population size.

D20SY07PS1036

AN EXPERIMENTAL TEST OF A POTENTIAL POST-COPULATORY PRE-ZYGOTIC REPRODUCTIVE BARRIER IN A PASSERINE SPECIES PAIR

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Sexual selection may drive speciation, but most research has focused only on pre-copulatory sexual selection, overlooking post-copulatory processes. Under strong post-copulatory sexual selection, postcopulatory pre-zygotic (PCPZ) phenotypes could diverge, limiting gene flow upon secondary contact. We did in vitro experiments on a potential PCPZ barrier between sister species: house sparrows (Passer domesticus) and Spanish sparrows (P. hispaniolensis). In birds, sperm selection most likely occurs as sperm cross the vagina, so we tested if reproductive tract fluid of heterospecific females reduced sperm swimming speed or motility, relative to conspecific female fluid. We found that house sparrow female fluids affected the two species' sperm asymmetrically, consistent with the observed asymmetrical genetics of the hybrid species, the Italian sparrow (P. italiae, which has house sparrow mitochondrial DNA but mixed house and Spanish sparrow nuclear DNA). In house sparrow female fluid, slow-swimming house sparrow sperm were sped up, and fast-swimming house sparrow sperm were slowed down. Spanish sparrow sperm was not systematically affected, thereby increasing the speed difference between fast-swimming Spanish sparrow sperm and randomly-chosen house sparrow sperm. This could contribute to asymmetrical introgression if Spanish sparrow males with fastswimming sperm are also more likely to seek extra-pair copulations, which is plausible. Overall, however, there was no evidence for a PCPZ barrier: sperm performed equally in conspecific and heterospecific female fluids, and the species had similar sperm morphology, sperm swimming performance, and female fluid protein profiles. Low divergence in PCPZ phenotypes between species may be insufficient to cause an overall PCPZ barrier, and may be due to low post-copulatory sexual selection within each species. Also, pre-copulatory barriers between the species may prevent PCPZ barriers from evolving via reinforcement.

D20SY07PS1082

ECOLOGICAL AND MOLECULAR CUES FOR COLOUR CHANGE IN A MARINE FISH

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Coral reef fish are among the most colourful and beautifully patterned organisms on the planet; however, understanding the function and evolution of such coloured visual signals is unclear. Possible ways in which we can start to unlock the processes that have created such diversity in colours and patterns is to understand: 1) the molecular capacities that allow signal emergence, 2) how such visual signals are perceived by others (predators, competitors and potential mates), and 3) how this drives the evolution of such signals. To address these questions we are currently using the coral reef fish Pseudochromis fuscus as a model system. It is a species that exhibits multiple colour morphs, has the ability to change colouration between morphs, and is considered a putative aggressive mimic. However, the environmental cues that drive and the molecular basis that enable the species to change colour remain unclear. To investigate these questions we are using a multidisciplinary approach by combining behavioural, neurophysiological, cell histological and molecular methodologies. First results indicate mimicry as the main trigger for colour change in Pseudochromis fuscus. However, other benefits such as cryptic and predatory advantages might be associated with colour differences in this species. Ultimately we aim to better understand how colourful signals evolve, what kind of selective pressures might act on them and whether there are molecular similarities between the signal co-evolution of mimic and model species.

D20SY07PS1335

EFFECT OF WOLBACHIA INFECTION ON THE COURTSHIP SONG OF DROSOPHILA PAULISTORUM AND D. EQUINOXIALIS

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Understanding the processes that can lead to speciation are one of the main goals in evolutionary biology. It has been proposed that coevolution between parasite and its host can potentially drive speciation. A good model for studies of infectious speciation is the neotropical fly Drosophila *paulistorum spp.*. This species complex is currently under incipient speciation in nature and consists of six semispecies that are in an obligatory mutualistic relationship with bacteria of the Wolbachia genus. Previous studies have determined that infection with Wolbachia is associated with hybrid inviability and male sterility. Interesting enough, Wolbachia not only cause postzygotic isolation in the D. *paulistorum* complex but also prezygotic isolation through an influence on assortative mating between the different semispecies. These data suggest that the infection is modulating some courtship signal that allows self-recognition among the semispecies. Among the different male signals involved in mate recognition in *Drosophila* species, courtship song is one of the most studied and is well known to affect isolation between close related species. Here we recorded the courtship song of males from three D. paulistorum semispecies and also of its sibling species D. equinoxialis comparing wild type infected lines with lines treated with antibiotics where the bacteria titer were reduced. Our result shows a significant variation in the interpulse interval (IPI) associated with the infection with Wolbachia in all groups. We discuss whether the changes are likely to influence sexually selection or are more likely to reflect changes in male condition following curing of the Wolbachia infection.

Symposium

8. Evolutionary Significance of Epigenetic Variation

23 and 24 August



Program

Friday 23 August

Session(s): 12, 13 Saturday 24 August

Session(s): 14

Organisers: Oliver Bossdorf and Koen Verhoeven

Invited speakers: Frank Johannes, Carlos Herrera and Christina

Richards

Description:

Epigenetic mechanisms can have long-lasting effects on phenotypes, but what role do they play in evolution? Recent research provides evidence that epigenetic mechanisms can create heritable trait variation, that epigenetic variation can be heritably altered by the environment. Moreover, studies in natural populations suggest an epigenetic role in adaptation. This symposium presents the current status of the growing research field of ecological and evolutionary epigenetics.

8. Evolutionary Significance of Epigenetic Variation

D23SY08IT15:45R6

EPIGENETIC RESPONSE TO NOVEL AND CHANGING ENVIRONMENTS

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An essential component of deciphering the impact and long-term consequences of changing climatic conditions is understanding how organisms are able to respond to the environment. While studies interested in adaptation have focused on DNA sequence variation, and the assumption that trait variation is based on sequence variation, there is now pressing need to explore the role of epigenetic processes. Epigenetic effects can result in heritable, novel phenotypes even without variation in DNA sequence and could therefore provide an unappreciated source of response. The implications of epigenetic effects for the evolution of traits are just beginning to be explored, but epigenetic variation may expand the ecological and evolutionary options species in the face of rapid climate change. My lab group is exploring the potential role of epigenetic processes in natural and controlled studies of native, invasive and model plant species. Combined these studies should enhance our understanding of how epigenetic variation may be shaped by environment and contribute to adaptation.

D23SY08IT17:45R6

ECOLOGICAL EPIGENETICS OF NON-MODEL PLANTS: EXPLORING THE EVOLUTIONARY RELEVANCE OF EPIGENETIC VARIATION IN NATURAL PLANT POPULATIONS

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Epigenetic variation is often an important source of phenotypic variation across species and conspecific individuals of wild plants. In addition, intraspecific epigenetic variation frequently is spatially structured and correlated with biotic and abiotic environmental factors. The evolutionary relevance of natural epigenetic variation is, however, contingent on its being transgenerationally heritable and largely autonomous from genetic differences. It is therefore crucial to investigate these two key issues on wild plant populations. Fingerprinting phenotypically distinct individuals from different populations using both molecular epigenetic and conventional genetic markers may allow to determine whether genetic and epigenetic variation are coupled across sites and individuals or tend instead to be independent of each other. The alternation of generations characteristic of higher plants, whereby a succession of diploid sporophytes and haploid gametophytes take place in populations, allows to evaluate the extent to which molecular epigenetic markers are robust or not to epigenetic checkpoints operating at gametogenesis. The application of this research programme to wild populations of the perennial herb Helleborus foetidus in southern Spain revealed that variation across individuals in genetic (SSR and AFLP) and epigenetic (MSAP; methylation-sensitive amplified fragment polymorphism) markers were largely decoupled. Although the strong epigenetic differentiation between populations was in large part caused by MSAP markers that were reset at gametogenesis, there was still a sizeable proportion of markers robust to gametogenesis whose variation contributed significantly to epigenetic differentiation of populations. Epigenetic differentiation of *H. foetidus* populations seems to reflect local adaptation to the variable environment mediated by an epigenetic inheritance system.

D24SY08IT10:30R6

MAPPING THE TRANSGENERATIONAL EPIGENETIC BASIS OF COMPLEX TRAITS IN ARABIDOPSIS

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Quantifying the impact of heritable epigenetic variation on complex traits is an emerging challenge in population biology. Here we analyzed a panel of nearly isogenic Arabidopsis lines which segregate experimentally induced DNA methylation changes genome-wide. We provide compelling evidence that a small number of transgenerationally stable differentially methylated regions (DMRs) act as bone fide epigenetic quantitative trait loci (QTL^epi) in this population, accounting for 60-90% of the observed heritability underlying two complex traits, flowering time and root length. We show that these QTL^epi are reproducible and can be subjected to artificial selection. Over 75% of the putative causal DMRs within the QTL interval are also variable in wild populations of this species and are not significantly associated with cis or trans acting SNPs. These sequence-independent DMRs may be an important source of phenotypic diversity in ecological settings and thus provide a basis for Darwinian evolution.

8. Evolutionary Significance of Epigenetic Variation

D23SY08RT16:33R6

EPIGENETIC INHERITANCE IN INVASIVE SPECIES

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Epigenetic variation is one causal mechanism for phenotypic variation, and epigenetic rearrangements are thought to be involved in many cases of adaptive phenotypic plasticity. In environments that are temporally autocorrelated the phenotype of successfully reproducing individuals, and thus their epigenetic state, is predictive of the selective environment that will face their offspring. Some degree of epigenetic inheritance may therefore be beneficial in variable environments. Assuming that transmission of epigenetic markers is under genetic control we develop a model to explore the patterns of epigenetic inheritance in an organism that invades previously uncolonized and spatially variable areas. We find that the optimal degree of epigenetic inheritance does indeed vary across the invasion front: epigenetic transmission tends to be less faithful at the front than in areas that have been colonized for longer, where more stable epialleles may be the norm. We relate our results to the spatial structure of the environment and the dispersal kernel of the organism.

D23SY08RT16:57R6

INVESTIGATING THE CONTRIBUTION OF EPIMUTATIONS TO LONG-TERM ADAPTATION USING MODELS AND EXPERIMENTS

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There is more to heredity than DNA sequence alone. Epigenetic changes are defined as changes in gene expression due to chromatin modifications without changes in DNA sequence. We know that some epigenetic changes can be transmitted between generations. Epimutations can be encoded by variety of mechanisms including histone modifications, changes in methylation patterns, or even small RNAs. However, investigations of how transgenerational epigenetic inheritance may affect adaptation are only just beginning. We use a combination of modeling and experimental microbial evolution to investigate how transgenerational epigenetic inheritance affects adaptive walks, where fitness increases in a population by the sequential fixation of novel beneficial genetic mutations (or novel beneficial epimutations as well). In our model, we address how different assumptions about how the epigenetic system works affect its role in adaptation. In particular, we contrast the evolutionary effects of heritable epigenetic changes that are genetically-encoded responses to environmental change (adaptive plastic responses that can be transmitted) with epigenetic variation is sequence independent and random with respect to fitness (pure epigenetic variation). We find that epimutations can contribute to adaptation alongside genetic mutations if they are reasonably stable, and outline cases where epimutations can speed up adaptation relative to an equal mutational supply of genetic mutations, and cases where genetic assimilation is likely (or unlikely) to happen. We test the predictions of our model using experimental evolution with the unicellular algae Chlamydomonas reinhardtii. By using different selective environments and manipulating the epigenetic system both chemically and genetically, we show how differences in epimutation supply affect fitness gain over approximately 150 generations.

D23SY08RT18:33R6

SELECTION ON EPIGENETIC VARIATION MAY HAVE BEEN IMPORTANT IN DOMESTICATION OF CHICKENS

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Epigenetic variation may cause broad phenotypic effects in animals. However, it has been debated to what extent expression variation and epigenetic modifications, such as patterns of DNA methylation, are transferred across generations, and therefore it is uncertain what role epigenetic variation may play in evolutionary processes. We compared gene expression and methylation profiles in thalamus/hypothalamus in Red Junglefowl, the ancestor of domestic chickens, and a domesticated egg laying breed (White Leghorn, WL). There were significant differeces in gene expression as well as methylation, which were largely maintained in the offspring, demonstrating reliable inheritance of epigenetic variation. More than 70% of the differentially methylated loci were hypermethylated in WL, indicating that methylations have accumulated during domestication. Furthermore, there was an overrepresentation of differentially expressed and methylated genes in selective sweep regions, previously shown to be associated with chicken domestication. The results show that epigenetic variation is inherited in chickens, and we suggest that selection of favourable epigenomes, may have been an important aspect of chicken domestication. This could have happened either by selection of genotypes affecting epigenetic states, or by selection of methylation states which are inherited independently of sequence differences. The relationship between specific epigenetic variants and phenotype remains to be investigated.

D23SY08RT18:57R6

ENVIRONMENTALLY INDUCED EPIGENETIC PLASTICITY IN THE HUMAN PARASITE SCHISTOSOMA MANSONI

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Adaptation to environmental changes is based on the perpetual generation of new phenotypes. We know that phenotypic variability generating mechanisms have not only a genetic but also an epigenetic component, and their relative importance in adaptive evolution is an open question. Variability generating mechanisms are particularly important in host-parasite interaction models in which selective pressures are high and evolution is fast. Epigenetics has been proposed to be the language that is used to communicate between genome and environment. We present here data for a metazoan parasite/host system. The digenetic trematode Schistosoma mansoni is a human parasite that uses the mollusc Biomphalaria glabrata as intermediate host and humans as definitive host. We exposed S. mansoni populations to a stressful but ecological realistic environment: the interaction with an allopatric B. glabrata host in which the parasite develops into the human-infecting cercaria. We then studied phenotypic traits, epigenetic and transcriptional changes that the parasite engages in response to this stressful environment. ChIP-seq studies were performed with antibodies that recognize euchromatic and heterochromatic marks on cercariae released either from the allopatric or sympatric hosts. Differences in chromatin structures were found in roughly 0.1% of the epigenome (excluding repetitive sequences). RNA-seq studies performed on the same stages allowed the identification of about 200 differentially expressed genes between the stressful and normal conditions. Among those we indentified a histone-methyltransferase, providing a potential functional link between stress induced transcriptional and epigenetic modifications. More importantly, our data suggest that chromatin structure changes during the development of the cercariae are inherited by the subsequently formed adult stage.

D24SY08RT11:18R6

ADAPTIVE DYNAMICS MODELLING WITH EVOLVING EPIGENETICS

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The term epigenetics is used to cover both the development of phenotypes from genotypes and the existence of alternative epigenetic birth states for a given genotype. If we define genotype-phenotype maps very broadly as the phenotypes and phenotype distributions a genotype can produce, then we can use these to model how an evolutionary dynamics depends on underlying traits and genotypic variation which control apparent phenotype distributions. Such models can then investigate different epigenetic mechanisms simultaneously evolving. A long-term perspective on fitness and evolutionary dynamics is essential to understand whether an epigenetic architecture is adaptive. I propose to use invasion fitness of mutants and adaptive dynamics approximations to investigate this. For such approximations in models with apparent and underlying traits of individual phenotypes and alleles, expressions for fitness gradients and evolutionary stability criteria are derived. I complement these with tools to investigate models where the apparent traits are life history fitness components such as stage-specific survival probabilities, and probabilities that gametes switch from the epigenetic states of their parental alleles at a certain locus into another. It is shown that a per-generation perspective on the dynamics of populations of mutants is essential to get good insight in the evolutionary dynamics of alternative epigenetic birth states. A specific example with evolving plastic epigenetic switching and evolving juvenile survival probabilities is worked out to discuss how trans-generational effects of parental birth state could evolve in a given ecological setting. I pay particular attention to the existence of alternative evolutionary outcomes for the same ecology, and to whether randomizing or plastic epigenetic switching strategies are most adaptive.

D24SY08RT11:42R6

EVOLUTION WITH STOCHASTIC EPIGENETIC VARIATION: A ROLE FOR RECOMBINATION

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Phenotypic adaptation to fluctuating environments has been an important focus in the population genetic literature. Previous studies have shown that evolution under temporal variation is determined not only by expected fitness in a given generation, but also by the degree of variation in fitness over generations; in an uncertain environment, alleles that increase the geometric mean fitness can invade a randomly mating population at equilibrium. This geometric mean principle governs the evolutionary interplay of genes controlling mean phenotype and genes controlling phenotypic variation, such as genetic regulators of the epigenetic machinery. Thus, it establishes an important role for stochastic epigenetic variation in adaptation to fluctuating environments: by modifying the geometric mean fitness, variance-modifying genes can change the course of evolution and determine the long-term trajectory of the evolving system. The role of phenotypic variance has previously been studied in systems in which the only driving force is natural selection, and there is no recombination between mean- and variance-modifying genes. We have developed a population genetic model to investigate the effect of recombination between mean- and variance-modifiers of phenotype on the geometric mean principle under different environmental regimes and fitness landscapes. We show that interactions of recombination with stochastic epigenetic variation and environmental fluctuations can give rise to complex evolutionary dynamics that differ from those in systems with no recombination.

POSTERS

8. Evolutionary Significance of Epigenetic Variation

D23SY08PS0694

THE ENVIRONMENT AS A SOURCE OF EPIGENETIC VARIATIONS

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Asexual lineages may have a widespread distribution in absence of genetic variation. Epigenetic processes have been proposed as a mechanism enabling such a response to environmental heterogeneity and thus represent an alternative mechanism to explain the ecological success of genetically identical organisms. This study aims to determine whether epigenetics reflect how asexual organisms deal with environment heterogeneity. We compared epigenetic profiles of sympatric lineages of the clonal fish *Chrosomus eos-neogaeus* from lakes of the north-eastern North America. While individual epigenetic differences represent 20% of the observed variation, individuals of a given lineage within a lake cluster together. Sympatric lineages share numerous epigenetic modifications and "lake effect" account for more than 40% of the variation, suggesting similar response to environment. Similarly, a given lineage from different environments displays numerous epigenetic differences. Finally, genetic differences also contribute to an important part of variation. This may be explained by two non exclusive hypotheses: response to environment differs according to lineage and/or sympatric lineages coexist in different ecological niches. Altogether, theses results suggest a strong role of epigenetics when dealing with different environments in absence of genetic variation.

D23SY08PS1027

BEHAVIOURAL EPIGENETICS AND THE EFFECTS OF DOMESTICATION IN THE CHICKEN

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Environmental changes and selection puts pressure on an organism's ability to adapt to new settings. An example where accelerated evolution in a short time-span has generated a large variety of phenotypes is seen in domestication, where artificial selection for desired traits have driven diversity not just from the founding origin but also in a range of different directions. Among the proposed explanations for this rapid change in phenotypes are epigenetic mechanisms. These are not only more frequent and flexible than genomic mutations, but their potential to shape individuals as well as their offspring make them viable targets for investigating the effects of environmental conditions or stimuli on an organism. With the chicken (Gallus gallus) as our model organism, we have been attempting to not only characterize behavioural differences between domestic chickens and their wild ancestors, but also specify genetic and epigenetic changes by looking at mutations, gene expression and epigenetic markers such as DNA methylation. Besides clear behaviour differences, we have found correlational changes between methylation patterns and gene expression, along with an enrichment of methylation in domestic chicken promoters. The genomic and epigenomic background, along with behavioural aspects, are currently being investigated in several projects including the effects of early stress, and an artificial selection line. Our hope is that the information generated from our experiments will give us insight in the stability and transmission of epigenetic markers, and let us expand the field of behavioural epigenetics. With knowledge about epigenetic changes in domestic animals, our understanding of their susceptibility to environmental changes such as stress or nutrition may help us in increasing animal welfare for both poultry and livestock.

D23SY08PS1383

A GENOME-WIDE COMPARATIVE STUDY OF DNA METHYLATION IN GREAT APES

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It has been hypothesized that differences between humans and their closet relatives may be explained by changes in gene regulation rather in primary genome sequence. Epigenetic alterations are involved in many biological processes and have been under-explored in comparative genomics. Specifically, DNA methylation is still poorly understood in the context of recent human evolution. In this study, we performed a comparative analysis of CpG methylation patterns between 9 humans and 23 primates including all four species of great apes (chimpanzee, bonobo, gorilla and orangutan) using Illumina Methylation450 bead arrays. Using this approach, we were able to study the dynamics of DNA methylation and to identify regions showing species-specific methylation pattern among the great apes, including ~130 genes with a pattern unique to human. We also identified a significant positive relationship between the rate of coding variation and alterations of methylation at the promoter level, indicative of co-occurrence between evolution of protein sequence and gene regulation.

8. Evolutionary Significance of Epigenetic Variation

D23SY08PS1396

CAN EPIGENETIC DIFFERENTIATION CAUSE THE FORMATION OF ECOTYPES? INSIGHTS FROM STABLE ALTITUDINAL VARIANTS IN THE MOUNTAIN PLANT HELIOSPERMA PUSILLUM (CARYOPHYLLACEAE)

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Adaptation is a process which continuously moves populations towards better fit phenotypes. In its widest sense, it involves short-term effects such as mechanisms of short-term transcription regulation, and long term-effects which result from natural selection acting on different kinds of heritable variation (i.e. segregating [epi-]allelic variants). Variations in biotic and abiotic conditions thus often lead to the formation of "ecotypes", i.e. distinct populations adapted to their specific habitat. If gene flow is limited, initial inter-fertility with other conspecific ecotypes may change into increasing divergence and incompatibility over time, and eventually result in speciation. An interesting example for multiple, independent origins of ecotypes is found in the mountain plant Heliosperma pusillum s.l., which comprises a widespread alpine ecotype inhabiting mountain creek banks and moist calcareous screes (*H. pusillum* s. str.), as well as a lowland ecotype with disjunct and locally limited distribution growing in gorges and under overhanging cliffs (*H. veselskyi*). AFLP fingerprints as well as non-coding chloroplast and nuclear sequences consistently indicate that there is no genome-wide genetic differentiation between these altitudinal variants which would mirror the conspicuous morphological and ecological differences. Nevertheless, morphology of the two types remains stable for at least one generation in offspring grown from seeds of high- and low altitude accessions in a common garden. We use methylation sensitive amplified polymorphism (MSAP) to test for genome-wide differences in DNA methylation in six pairs of high- and low altitude ecotypes from the Eastern Alps. We discuss the correlation of methylation patterns with evident phenotypic differences and the possible role of epigenetics in the initial phase of divergent evolution of ecotypes.

Symposium

9. Genetics and Genomics of Host-Parasite Coevolution

21 and 22 August



Program

Wednesday 21 August

Session(s): 5, 6, 7, 8

Thursday 22 August

Session(s): 9

Organisers: Joachim Kurtz and Hinrich Schulenburg

Invited speakers: Francis Jiggins and Ellen Decaestecker

Description:

Host-parasite coevolution often leads to fast evolutionary changes. It thus represents an ideal model for studying processes of rapid adaptation. Key to these studies is an understanding of the underlying genetics and genomics, because they help us identify the specific traits under selection and also the type of selective processes involved. Our symposium focuses on current advances in this field, taking advantage of recent innovations in high throughput sequencing technology.

D21SY09IT10:30R4

THE EVOLUTION AND GENETICS OF SUSCEPTIBILITY TO INFECTIOUS DISEASE

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It is common to find considerable genetic variation in susceptibility to infection in natural populations. Not only is this variation abundant, but it can have important implications for health, economic output and the coevolution between hosts and parasites. Within populations of Drosophila, we have found that selection by viruses tends in increase genetic variation in disease susceptibility, due to directional selection increasing the frequency of recently arisen major-effect alleles that increase resistance. Furthermore, bacterial symbionts can also protect many species of Drosophila against viral infection, and the spread of these symbionts also has the potential to generate rapid shifts in the susceptibility of flies to viruses. Overall, susceptibility to viral infection is controlled by a small number of major-effect genes and symbionts that are under strong selection and rapidly change in frequency.

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HOST ALLELIC DIVERSITY DRIVES LONG-TERM HOST-PARASITE COEVOLUTIONARY DYNAMICS

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Background: Hosts and parasites are involved in a coevolutionary interaction in which hosts do not evolve as fast as their parasites. Yet, fast adaptive genetic changes occur upon infection, especially if host-parasite interactions are characterized by Red Queen dynamics. Red Queen dynamics between both antagonists are caused by negative frequency-dependent selection and are assumed to have constant amplitudes. Here, a long-term time shift experiment, based on a unique historical reconstruction of a *Daphnia*-parasite coevolution, reveals that infectivity cycles with a smaller amplitude in experienced than in naive hosts. Experienced hosts were isolated from recent time periods, naive hosts from past time periods. A coevolution model, incorporating an increase in allelic diversity over time in the host confirmed the asymmetry in the infectivity cycles. In contrast, increased virulence over time did not confirm the observed experimental results. The accumulation of resistance alleles affects long-term Red Queen dynamics. Long-term effects in host-parasite coevolution have so far been neglected, but this reconstruction in combination with a theoretical study on long-term time shifts between a host and a parasite extends current insight into the dynamics of co-evolutionary antagonistic interactions.

9. Genetics and Genomics of Host-Parasite Coevolution

D21SY09RT11:18R4

EXPERIMENTAL EVOLUTION FOR INCREASED VIRUS RESISTANCE IN DROSOPHILA MELANOGASTER REVEALS NO COSTS AND A SIMPLE GENETIC BASIS

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Because hosts and parasites exert strong selection pressure on each other, it is particularly relevant to study their interaction in an evolutionary context. Experimental Evolution permits the establishment of causality between evolutionary processes and adaptation patterns. Here we use experimental evolution of Drosophila melanogaster exposed to Drosophila C virus (DCV) to address the phenotypic and genotypic changes of hosts evolving in presence of parasites. Upon exposure to the virus, Drosophila survival increased from 33% to almost 90% after 35 generations of selection. This response carried no detectable costs in fitness in the absence of infection, and was not lost after 10 generations in the absence of selection. Cross-resistance was found for other viruses, such as CrPV and FHV, but not to bacterial pathogens. Whole genome sequencing of pooled samples of virus-selected populations and their matching controls at generation 20 uncovered two regions of significant differentiation between these groups of populations. The first corresponded to a region of 4 megabases(Mb) in the 3L chromosomal arm. This region's peak of differentiation corresponded to a polymorphism in *pastrel* (pst), a gene recently associated with increased DCV resistance. The second was a pair of significantly differentiated SNPs in the X chromosome, in genes not previously associated with virus resistance. Results with a panel of deficiencies in the 3L chromosome confirmed that deficiencies which encompass *pst* are the ones with more influence on survival after DCV infection, in a region of approximately 2 Mb. There is ongoing work to confirm the involvement of other candidate genes in this region and of the genes in the X chromosome in resistance to DCV infection. Overall we show that selection for increased virus resistance I) is stable and bears little costs, II) is advantageous in the defense against other viral pathogens, and III) has a simple genetic basis.

D21SY09RT11:42R4

DISCOVERY, DISTRIBUTION AND EVOLUTIONARY GENOMICS OF VIRUSES NATURALLY INFECTING DROSOPHILA MELANOGASTER

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Drosophila melanogaster is an important model for innate immunity, and is arguably our primary model for antiviral resistance in arthropods. Several groups have used population-genetic and phylogenetic approaches to show that some antiviral immune genes in *Drosophila* (notably the antiviral RNAi pathway) display highly elevated rates of adaptive evolution. However, although this is consistent with a host-virus arms race, the evolutionary genetics of Drosophila viruses are almost unstudied - only a handful of viruses which naturally infect *Drosophila melanogaster* are known, and only Drosophila Sigma Virus (a Rhabdovirus) has been regularly isolated from wild populations. In an attempt to understand the evolutionary genetics of Drosophila viruses, we have sequenced both RNAseq, and small-RNA, libraries from large pooled samples of wild-caught D. melanogaster. This has allowed us to identify several new viruses, including several RNA viruses (viruses with sequence similarity to Sacbrood Virus, Slow Bee Paralysis Virus, Chronic Bee paralysis virus, Acyrthosiphon Pisum Virus, Flaviviruses, and Cypoviruses) and a DNA virus (Nudivirus). Following a geographic survey of *D. melanogaster*, we find that the previously known viruses of *D. melanogaster* (including DAV, Sigma and Nora) are widespread at low to intermediate prevalence. None of the viruses shows high rates of adaptive evolution, and in general (despite substantial synonymous divergence) protein sequences are very highly conserved. However, while this may indicate that these viruses are not engaged in 'arms race'-like coevolution, we suspect that the short timescale of viral co-ancestry (tens to hundreds, rather than thousands, of years) makes this process extremely difficult to detect. This is in sharp contrast to viral evolution in response to vertebrate adaptive immunity, which adapts plastically on the same timescale as viral evolution.

9. Genetics and Genomics of Host-Parasite Coevolution

D21SY09RT14:00R4

PARASITOID RESISTANCE IN DROSOPHILA: FROM SHORT TO LONG TERM EVOLUTIONARY PATTERNS

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With the availability of genome sequences of closely related species and the tools to investigate genome changes, we have come closer to understand the complexity of the evolutionary history of immune systems. We investigate the genomic changes associated with the evolution of cellular immunity in the *Drosophila* genus, focusing on the ability to encapsulate and melanize parasitoid eggs. While parasitoid wasps are widely distributed and a common threat for many insects, within the Drosophila genus there is large variation in encapsulation ability, from completely absent in some species to high resistance in others. Through a combination of parasitization-experiments and comparative genomics we found that the ability to encapsulate parasitoid eggs has evolved in a branch of the *Drosophila* lineage and has subsequently been lost in one species of this group. The evolution of this trait is associated with the acquiring of a special type of differentiated blood cells, the lamellocytes. Some key genes that play important roles in the cellular immune defense are found to be restricted to this taxonomic group, and show large-scale sequence changes in the species that lost the ability to resist against parasitoids. Variation in resistance is not only present at the inter-species level but also between populations inside one species. Understanding the source of this variation requires a better understanding of the mapping from the genome sequences to the phenotypic pattern. For this, we are currently comparing the transcriptome (RNAseq) after parasitization with the wasp Asobara tabida of 4 species and 2 artificially selected lines for increase resistance. This set of data will allow us to contrast the long term variation found between species with the short term evolution found in the selected lines.

D21SY09RT14:24R4

GENOMIC ADAPTATION IN THE BRACOVIRUS OF COTESIA SESAMIAE IDENTIFIED BY TARGETED RESEQUENCING

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Cotesia sesamiae are small parasitoid wasps parasitizing over twenty lepidopteran African stem borer species. It is thought that local adaptation to these different host species is mediated by their symbiotic bracoviruses (BVs). BVs derive from large DNA viruses and have been stably integrated in the wasp genome. The wasp use BVs to introduce ~150 genes in parasitized caterpillars, inducing immunosuppression and allowing wasp larval development. In C. sesamiae, different alleles of the *CrV1* BV gene explain parasitic success in a particular host species. Nonetheless, other BV genes could be involved in wasp local adaptation or specialization to their lepidopteran hosts. To investigate this, we focused on the BV genome of 25 samples representative of different African C. sesamiae populations. As we worked with tiny organisms, we used custom-made targeted sequence capture to enrich our sequence library in BV genomes (257 kb) prior to Illumina resequencing. This proved to be a very efficient technique as we obtained high target coverage (1100X) and high percentage of mapped reads (90%) for all C. sesamiae populations and for the more distant outgroups. First, we used population genetics tools to identify regions under strong divergent selection by comparing nucleotide diversity (π) and genetic differentiation (FST) along the BV genome and between populations. Secondly, we used a phylogenomic approach to establish the evolutionary relationships between these populations using BV sequence of three outgroup species. Third, comparative genomics helped to assess the effects of particular mutations and identify sites evolving under positive selection. Last, we compared the molecular evolution of all orthologous BV genes both between and within populations and measured the rate of non-synonymous versus synonymous substitutions under a branch-site evolutionary model. Overall our results indicate that different BV genes are at play depending on local host context.

D21SY09RT14:48R4

IDENTIFICATION OF A MAJOR NATURAL POLYMORPHISM RESPONSIBLE FOR THE SENSITIVITY TO THE ORSAY VIRUS IN CAENORHABDITIS ELEGANS

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The nematode *Caenorhabditis elegans* and its natural pathogen, the positive-strand RNA virus Orsay have recently emerged as a new animal model of host-virus interaction. When infected, sensitive strains of C. elegans show disorganization of intestinal cells. The virus was originally detected in the C. elegans isolate JU1580 and is horizontally transmitted. Using a genome-wide association study on 97 wild *C. elegans isolates* and finer mapping with Near Isogenic Lines, we identified a region of 155 kb in the center of chromosome IV as carrying the major determinant of viral sensitivity. In JU1580, this region contains a 159 base-pair deletion called *niDf*250, affecting the conserved *drh-1* gene (encoding a RIG-I-like helicase). We found that DRH-1 is required for the initiation of an antiviral RNAi pathway and the generation of virus-derived siRNAs (viRNAs). In mammals, RIG-I-domain-containing proteins trigger an interferon-based innate immunity pathway in response to RNA virus infection. Our work in *C. elegans* demonstrates that the RIG-I domain has an ancient role in viral recognition. We propose that RIG-I acts as modular viral recognition factor that can couple viral recognition to different effector pathways including RNAi and interferon responses. Surprisingly, the *drh-1* deletion - the derived sensitive allele - is commonly found in *C. elegans* wild populations (23% of wild isolates). Two main hypotheses could explain this distribution: 1) *niDf*250 is linked to a beneficial allele or 2) *niDf*250 is beneficial by itself in certain conditions. Supporting the first hypothesis, *niDf250* lies in a region with strong linkage disequilibrium. To further test these hypotheses, we are now performing laboratory fitness and competition experiments to decipher the impact of the *niDf*250 allele and the linked region on *C. elegans* fitness.

D21SY09RT15:12R4

MATRYOSHKA HYBRID ZONES: THE PINWORM SYPHACIA OBVELATA IN THE EUROPEAN HOUSE MOUSE HYBRID ZONE

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Hybrid zone studies of host-parasite coevolution have largely taken a host-centric viewpoint: does parasitism affect the outcome of host hybridization by differentially impacting the fitness of host taxa and their hybrid descendants? This viewpoint neglects the possibility that host taxa have distinctive genetic clusters of parasites that may themselves potentially hybridize. Hybridization, bringing together combinations of genes previously untested by natural selection, may affect parasites in similar ways to free-living organisms: revealing a barrier to gene flow, promoting divergence via reinforcement, homogenizing genetic clusters or leading to rapid adaptive diversification via the formation of hybrid parasite species. We use the European house mouse hybrid zone (HMHZ) between Mus m. musculus and M. m. domesticus and their pinworms to explore these issues. First, from a host-centric viewpoint, we tested whether hybrids have greater or lesser load of pinworms than additive expectations. Sampling of 689 mice from 107 localities typed for 1401 diagnostic SNPs across the Bavaria-Bohemia region of the HMHZ we found hybrids have significantly reduced pinworm load, contradicting the idea that pinworms reduce host hybrid fitness through increased load. Second, from a parasite-centric viewpoint, we analyzed the genetic structure of Syphacia obvelata across the same part of the HMHZ by typing 267 female worms from mice at 87 HZ localities and 18 allopatric localities for 10 microsatellite markers and the mtCOX1 gene. We found distinct genetic clusters of S. obvelata meeting at the host HZ, with hybrid pinworm genotypes at the centre. Our results are consistent with independent host-parasite arms races running during isolation of the host taxa: any accumulated coadaptation will breakdown on hybridisation of host and of parasite. The low parasite loads observed in the HMHZ centre may be due to "immune vigor" of hybrid hosts and/or low fitness of their hybrid parasites.

D21SY09RT15:45R4

USING RAD-SEQ AND CANDIDATE GENE SEQUENCING TO EXPLORE PATTERNS OF HETEROZYGOSITY AT DISEASE RESISTANCE GENES ACROSS NATURALLY INBRED AND OUTCROSSED PLANT POPULATIONS

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Models of plant resistance (R) gene evolution suggest a significant role for balancing selection in maintaining high levels of genetic diversity. Selection can then act on this pool of resistance gene alleles to allow plant populations to recognise a diverse range of pathogen genotypes. However, the ecology and life history of a species can have strong effects on patterns of genetic diversity across the genome, which may counter the maintenance of genetic variation through selection. In particular, intraspecific variation in mating systems can produce naturally outcrossed and inbred populations, with reduced heterozygosity at neutral genetic loci in inbred populations. This reduction in heterozygosity is expected to occur throughout the genome, including at genes important for pathogen recognition and resistance, which may reduce the potential for rapid evolution of pathogen resistance in inbred populations. The perennial plant, Arabidopsis lyrata, displays significant variation in mating system around the North American Great Lakes resulting in naturally outcrossed and inbred populations. Using Restriction Associated DNA (RAD) sequencing, we have genotyped 4 individuals from each of six inbreeding and six outcrossing populations of *A. lyrata* at ~ 30000 RAD loci. Using these sequence data, we can test for reduced heterozygosity across the genomes of individuals from inbred populations. We can also use these sequence data to estimate genetic diversity at or near candidate disease resistance genes (using comparisons to the well-annotated Arabidopsis thaliana genome). These analyses will be combined with sequence data produced via PCR amplification of particular resistance genes thought to be involved in the recognition of different pathogens of the Brassicaceae. Together these data should significantly improve our understanding of the genomic consequences of shifts in mating system and its potential effects on the evolution of disease resistance.

D21SY09RT16:09R4

COEVOLUTION AND LIFE CYCLE SPECIALIZATION OF PLANT CELL WALL DEGRADING ENZYMES IN A HEMIBIOTROPHIC PATHOGEN

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Zymoseptoria tritici is an important fungal pathogen on wheat that originated in the Fertile Crescent. Its closely related sister species Z. pseudotritici and Z. ardabiliae infect wild grasses in the same region. This recently emerged host-pathogen system provides a rare opportunity to investigate the evolutionary processes shaping the genome of an emerging pathogen. Here, we investigate genetic signatures in plant cell wall degrading enzymes (PCWDEs) that are likely affected by or driving coevolution in plant-pathogen systems. We hypothesize four main evolutionary scenarios and combine comparative genomics, transcriptomics and selection analyses to assign the majority of PCWDEs in Z. tritici to one of these scenarios. We found widespread differential transcription among different members of the same gene family, challenging the idea of functional redundancy and suggesting instead that specialized enzymatic activity occurs during different stages of the pathogen life-cycle. We also find that natural selection has significantly affected at least 19 of the 48 identified PCWDEs. The majority of genes showed signatures of purifying selection, typical for the scenario of conserved substrate optimization. However, six genes showed diversifying selection that could be attributed to either host adaptation or host evasion. This study provides a powerful framework to better understand the roles played by different members of multi-gene families and to determine which genes are the most appropriate targets for wet lab experimentation, for example to elucidate enzymatic function during relevant phases of a pathogen's life-cycle.

D21SY09RT16:33R4

IN SEARCH OF THE FUNGAL COMPOUNDS THAT MANIPULATE ANIMAL HOST BEHAVIOR

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Parasite-host co-evolution has driven parasites to evolve strategies to invade, overcome the immune system, and exploit their hosts for their own survival and dispersal. Some parasites also interact with the host's nervous system, changing behavior. One of the most dramatic examples is the fungus Ophiocordyceps unilateralis s.l. infecting Camponotus species, where ants bite into vegetation before dying to facilitate spore dispersal. To establish this, the fungus not only overcomes the immune system, but also manipulates the brain and atrophies the muscles. Through extensive fieldwork on three continents we established that a high diversity of manipulation phenomena involving ants and fungi exists. This provides a framework for comparative profiling of manipulator compounds using a combination of metabolomics and proteomics. We are combining metabolite and protein profiling with ex vivo insect tissue culturing to study compounds secreted in different areas of the host. Using this technique we established that generalist and specialist fungal entomopathogens react differently when presented with the same insect tissues. Next to that, these entomopathogens react heterogeneously to brain and muscle tissue by secreting a significantly different array of metabolites. Furthermore, O. unilateralis employs different metabolites when presented with brains from different ant species, while similar compounds are found as well. Identifying if the "behavioral manipulator" compounds are only found when the fungus is presented with its co-evolved host, or also when presented with other hosts, will inform us if host specificity lies at the level of behavioral manipulation or that of spore entry and overcoming the immune system. By combining metabolite and protein discovery we are identifying candidate compounds involved in manipulation. We are also testing the effect of potential brain manipulating compounds by combining artificial ant infection with behavioral assays.

D21SY09RT16:57R4

FUNCTIONAL ANALYSIS OF DIFFERENTIALLY EXPRESSED GENES IN THE SYMBIOTIC ASSOCIATION BETWEEN THE PILL-BUG ARMADILLIDIUM VULGARE AND THE FEMINISING WOLBACHIA

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Today, there is a wide consensus on the essential role of microbial associations to eukaryote evolution. In most cases, the relationship between host and symbiont is so close that the microorganisms cannot be cultured, making them difficult to study. However, high-throughput sequencing has offered new opportunities for symbiosis research. Due to their widespread association with the Wolbachia endosymbionts, terrestrial isopods represent a model system to understand intimate symbioses. Wolbachia are vertically transmitted facultative bacteria acting as reproductive parasites in isopods, inducing the feminisation of genetic males in the pill bug *Armadillidium vulgare*. Among the three feminising Wolbachia identified in this host, two strains (*w*VulC and *w*VulM) vary in their prevalence and extended phenotypes. wVulC, the most prevalent strain exhibiting the strongest feminising effect, is also the most virulent strain inducing various fitness costs. To decipher the conflicting associations between wVulC, wVulM and A. vulgare, we have constructed cDNA libraries from ovaries and from whole animals challenged by pathogenic intracellular bacteria according to their Wolbachia infection status. RNA from infected and uninfected animals was subjected to RNA-seq sequencing followed by *de novo* data assembly and annotation. This process generated a library of 33,120 annotated transcripts. Identification of differentially genes (DE) genes as well as overrepresented gene ontology (GO) terms was then carried out using the R packages DESeq and GOSeq. Interestingly the highest number of DE genes was recorded in the animals infected by the less virulent strain wVulM. In most treatments, these DE genes could be assigned to GO categories that are underrepresented when Wolbachia are on board. This study is part of the widest program ImmunSymbArt granted by the French National Research Agency which aims to determine the symbiotic syndrome in four model systems.

D21SY09RT18:33R4

POPULATION STRUCTURE OF MICROPARASITES INFECTING DAPHNIA: SPATIO-TEMPORAL DYNAMICS

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Surprisingly, in many field studies of natural host-parasite systems genetic changes have been investigated for one player only – the host. This is astonishing as coevolution requires changes in the frequencies of both players. The previous restrictions were mainly caused by limited access to molecular markers for unculturable microparasites. Recently, we have established a NGS protocol to shed light on the genetic changes within populations of *Caullerya mesnili* (Ichthyosporea), the parasite of the waterflea *Daphnia*. This parasite is one of the most common pathogens of European *Daphnia* species that inhabit large, permanent lakes. We show that the genetic structure of parasite populations varies both over space and, most interestingly, over time. Given its high virulence (up to 95% fecundity reduction), strong genetic specificity for infection, large prevalence in natural populations (up to 40%), and currently established molecular tools - *Caullerya* is a strong candidate to become a model parasite for future coevolutionary studies of natural host-parasite systems. Our preliminary studies show that the genetic structure of parasite populations varies both over space and, most interestingly, over times. In the near future, the new molecular and bioinformatical techniques will allow to track changes in genetic population structure on a large scale for the often unstudied member of a coevolutionary pair – the parasite.

D21SY09RT18:57R4

EVOLUTION OF DAPHNIA MAGNA RESISTANCE TO THE PATHOGEN PASTEURIA RAMOSA

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Crustaceans of the genus *Daphnia* have long been used as models in studies of ecology and evolution and of host-pathogen coevolution in particular. However, little is known about the genetic and molecular basis of Daphnia resistance to pathogens. A well-known ecology combined with recent advances in genomic, genetic and molecular tools make Daphnia crustaceans, and in particular Daphnia magna and Daphnia pulex, remarkable models for modern evolution and ecology. D. magna is colonized by a wide range of parasites and pathogens, among them the bacterium *Pasteuria ramosa*. The study of inheritance patterns of resistance and susceptibility of *D. magna* clones to different *P.* ramosa genetic isolates reveals strong genotype-to-genotype interactions, suggesting coevolution between host and pathogen populations. Resistance and susceptibility of *D. magna* to different *P.* ramosa isolates follow mendelian patterns of inheritance and segregation, suggesting that a small number of loci underlie natural variation in *D. magna* resistance to *P. ramosa* infection. A F2 panel of D. magna was generated, with more than 200 genotypes that are kept by clonal reproduction. A QTL analysis revealed one genomic region of 150kb in linkage group 4 that explaining approximately 60% of the observed variation. However, further inheritance analysis showed that at least 3 loci underlie natural variation in *D. magna* resistance to *P. ramosa* infection. One of those loci corresponds to an indel of approximately 50kb. We are currently fine-mapping the QTL interval to identify genes and polymorphisms underlying *Daphnia* resistance to *Pasteuria*. We aim to identify which genes and networks and which polymorphisms underlie the natural variation and evolution of pathogen resistance in *D. magna*. We plan to use molecular tools recently developed for *D. magna* to achieve our objectives.

D22SY09RT10:30R8

MALE PREGNANCY AND IMMUNE DEFENCE ANOMALY

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The major histocompatibility complex (MHC) - mediated adaptive immune system was claimed to be the hallmark of vertebrate immune defence. Now recent work suggests that vertebrate immune systems and in particular the MHC - mediated immunity are much more plastic than previously assumed. Based on deep-transcriptome sequencing, we report the loss of the MHC class II immune pathway in the sexrole reversed pipefish Syngnathus typhle. In contrast, MHC class I genes were present, and their diversity correlates with the efficiency of immunity and male mate choice behavior. We identify "sexrole reversal" as a potential selection factor for loss of MHC II and other surprising anomalies in the pipefish immune defence, and give an outlook of how this can affect host-parasite coevolution.

D22SY09RT10:54R8

FOOTPRINTS OF DIRECTIONAL SELECTION IN WILD POPULATIONS OF ATLANTIC SALMON: EVIDENCE FOR PARASITE-DRIVEN EVOLUTION?

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European populations of Atlantic salmon (Salmo salar) exhibit natural variance in susceptibility levels to the ectoparasite *Gyrodactylus salaris*, ranging from resistance to extreme susceptibility, and thus are a good model for studying the evolution of virulence and resistance. Advances in genome technologies provide new opportunities for obtaining a genome-scale view of the action of natural selection in wild populations. However, distinguishing the molecular signatures of genetic drift and environmentassociated selection may challenge the search for specific pathogen driven selection. We used a novel genome-scan analysis approach aimed at i) identifying signals of selection in salmon populations affected by genetic drift at varying levels; and ii) separating the potentially selected loci identified into those affected by pathogen (G. salaris)-driven selection and salinity-driven selection. 4631 single nucleotide polymorphisms (SNPs) were screened in 472 salmon individuals from 12 different north European salmon populations. We identified several genomic regions potentially affected exclusively by parasite-driven selection, as well as several regions affected by salinity mediated directional selection. Functional annotation of candidate SNPs supported the participation of the detected genomic regions in immune defense and osmoregulation. These results provide new insights in genetic basis of pathogen susceptibility/resistance and adaptation to various salinity levels in Atlantic salmon, and open possibilities for specific candidate gene search.

D22SY09RT11:18R8

CAN PARASITES DRIVE POPULATION DIVERGENCE IN THREE-SPINED STICKLEBACK?

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The role of parasites in driving the evolution of hosts is poorly understood. Adaptive evolution occurs with a change in adaptive allele frequencies derived either from standing genetic variation or from new mutations. Looking at population structure of adaptive genes together with neutral markers may reveal whether the adaptive traits that differ between populations are likely to have resulted from divergent selection or genetic drift. Parasites can be potent agents of selection which act both directly on survival and have also been known to affect mate choice. The three-spined stickleback Gasterosteus aculeatus is a good model system to study mechanisms of adaptive evolution due to its propensity to radiate into young postglacial habitats. We have surveyed the composition and abundance of parasites in stickleback populations on the Island of North Uist, Scotland for four years, and conducted infection experiments on lab bred naïve progeny. Our previous results show that populations display adaptation to their parasites in that they are resistant to naturally occurring parasites. Furthermore this is genetically based; maternal effects were ruled out through breeding schemes and experiment replicates over generations. An RNAseq experiment was conducted and used to identify genes that were differentially expressed when fish were experimentally infected with a parasite. Some of these genes were selected together with known candidate immune genes. Linked microsatellites were found and genotyped together with a set of neutral microsatellites (as controls). Stickleback samples were from populations with known parasite communities. Any detectable selection on these loci, together with adaptive (immune) and neutral structure will be presented in correlation with parasite composition.

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HOST-SPECIFICITY IN HYBRIDS OF TWO SCHISTOCEPHALUS SPECIES WITH DIFFERENT STICKLEBACK HOSTS

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Schistocephalus solidus is a highly specific tapeworm that only infects the three spined stickleback Gasterosteus aculeatus as a second intermediate host. The closely related S. pungitii uses the ninespined stickleback *Pungitius pungitius* as second intermediate host, showing the same host specificity at this level. Both parasites potentially share the same final hosts – piscivorous birds - and can occur in sympatry. It is therefore possible that natural hybridization takes place but they have not been detected. We used an in vitro breeding system to hybridize S. solidus and S. pungitii and quantified the hybridization rate using microsatellite markers. We measured several fitness relevant traits in pure lines of the parental parasite species as well as in their hybrids: hatching rates, infection rates in the copepod first host, and infection rates and growth in the two species of stickleback second hosts. We show that the parasites can hybridize in the in vitro system, although the proportion of self-fertilized offspring was higher in the heterospecific breeding pairs than in the control pure parental species. Hybrids have a lower hatching rate, but do not show any disadvantages in infection of copepods. In fish, hybrids were able to infect both stickleback species with equal frequency, whereas the pure lines were only able to infect their normal host species. This suggests that the hybrids have given up the host specificity and raises the question why natural hybrids have not been identified in the wild. We are currently investigating pre- and postzygotic mechanisms that might prevent natural hybridization. Our results furthermore indicate a co-dominance of the genes responsible for infection of different fish hosts. This system offers the unique possibility to investigate the genetic mechanisms underlying this host specificity using backcrosses and a OTL approach.

POSTERS

EVASION OR SUPPRESSION? INFECTION STRATEGIES OF A TRYPANOSOME GUT PARASITE OF BUMBLEBEES

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Parasites can either avoid host detection (evasion) or actively suppress host immune responses (suppression) in order to infect their hosts. These two strategies could have important consequences for multiple infections. Evasion would not be beneficial in mixed infection if the co-infecting strain fails to also evade, but immune suppression would benefit both infecting strains irrespective of the other co-infecting strain's strategy. Co-infection, in turn, provides the opportunity for both parasite competition and sexual recombination. The bumblebee *Bombus terrestris* is commonly infected by multiple strains of the trypanosome gut parasite *Crithidia bombi* in nature. We tested whether successful infection is achieved by suppressing or evading the host's immune system by experimentally infecting workers of several colonies with *Crithidia* strains that were either highly or lowly infective. We used both single and mixed inocula of *C. bombi* clones, where mixed infections consisted of pairings between a highly and a lowly infective strain. We measured host gene expression of 27 candidate genes and quantified infection intensities within the same individual 18 hours post infection. We discuss implications and insights into the complex genetics of host-parasite interactions.

LANDSCAPE GENOMIC ANALYSIS HIGHLIGHTS HOW PARASITE-DRIVEN PROCESSES AFFECT NEUTRAL AND ADAPTIVE GENETIC DIVERSITY IN RED GROUSE

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A major endeavour in evolutionary ecology is to elucidate the relative contribution of stochastic and deterministic processes to genetic diversity and how this influences fitness, population dynamics and adaptation. Genotyping individuals sampled densely from a heterogeneous landscape across both neutral markers and candidate genes considered to be functionally relevant for the ecological character of interest is a useful approach that informs an understanding of the interplay between ecological and evolutionary processes. The red grouse (Lagopus lagopus scoticus) is a Scottish upland game bird whose population dynamics are to a large degree affected by a nematode parasite (Trichostrongylus tenuis). Our aim is to shed light on the genetic basis of parasite response and adaptation, by employing a candidate gene approach on a landscape scale. We use 454 pyrosequencing to genotype 190 grouse sampled across north-east Scotland (Cairngorms area) at 12 candidate loci for parasite response (identified from previous microarray and sequence data), and 3 putatively neutral intronic loci. We identify associations between patterns in genetic variation and environmental variables, particularly landscape, parasite loads and management regimes. We further test for signatures of natural selection in genetic patterns, employing Fst outlier analysis with intronic loci as a neutral reference. Our results elucidate the influence of ecological dynamics on genetic diversity and the consequences for population dynamics in a management context for this economically important game bird species.

VARIABLE PATTERNS OF HOST SPECIALIZATION IN THE GENERALIST TICK IXODES RICINUS

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The degree of host specialization in parasites can greatly modify the nature and outcome of interspecific interactions. When parasites are also vectors, their ability to adapt to new hosts and their response to changes in host community structure will have important consequences for both their population dynamics and evolution, but may also cascade down to the microparasites they transmit. Although vector-borne diseases frequently involve vectors that can exploit several host species (i.e., generalists), little is known about their potential to specialize on locally available hosts and even less on the impact of specialization for pathogen transmission. A first step to better apprehend the importance of this phenomenon for the evolution, ecology and epidemiology of vector-borne disease systems is to study patterns of host-associated genetic divergence across diverse vector populations. We used this approach to study the host-vector-pathogen system involving the European tick *Ixodes* ricinus, its various vertebrate hosts and the bacteria responsible for Lyme disease (Borrelia burgdorferi s.l.). Significant levels of genetic structure among ticks of different host individuals and host types were found, but only within certain host communities. We predicted that longer established and more stable host communities would show stronger patterns of host-associated divergence than more recently colonised or perturbed communities. We tested this prediction by combining field samples from a European-wide transect that includes both the historical range of the tick species and its newly colonized zones. Our results reveal a complex pattern of parasite adaptation across this European landscape.

PARASITIC LIFESTYLE AND GENOMIC REDUCTION IN METAZOAN PARASITES

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Even though parasitism is the most common lifestyle on earth, the impact of the lifestyle on parasite genomes is still poorly known. Adaptation to a specialized niche has been shown to cause reduction of genome size in intracellular parasites (bacteria and eukaryotes) when compared to free-living organisms. However, this genomic reduction has been suggested to be connected to intracellular lifestyle only. We explored the effect of parasitism on genome size in four metazoan taxa: flatworms, nematodes, annelida and arthropods. We found that the genome size was significantly smaller in parasitic taxa when compared to closest free-living taxa in all the studied metazoans. Our results advocate that despite of the high variation in the taxonomic position, evolutionary history and diverse life cycles, parasitism as a lifestyle promotes genomic reduction. The selection for genomic reduction associated with parasitic lifestyle must be strong and adaptive. We suggest that small genome size benefits parasites by decreasing the costs of genome replication, thus increasing their growth and likelihood of successful transmission, leading to enhanced virulence. Because the number of sequenced metazoan parasite genomes is low, it is difficult to estimate if reduction in genome size is due to gene loss or by compaction, both of which are observed in protozoan parasites. In many protozoan parasites genome is reduced by loss of even complete core metabolic pathways, leaving the parasites parasitizing directly on the function of host genes. If similar findings are made with metazoan parasites, we suggest that parasitism on host genes, functions and metabolism could be considered as a core component of the definition of virulence (harm caused by the pathogen leading to reduction of host fitness).

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EXPERIMENTAL ADAPTATION OF THE PHYTOPHAGOUS SPIDER MITE, TETRANYCHUS URTICAE TO A NEW HOST PLANT

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Our ultimate goal is to identify the underlying genetic mechanisms of adaptation of phytophagous mites, *Tetranychus urticae*, to tomato plants, i.e. a new host species. For this purpose, we transferred in replicate lines on tomato plants individuals from two experimental groups consisting of independent ancestral populations previously maintained for four hundred generations on cucumber and bean plants. For each of the four (two ancestral and two evolved) treatments we measured on tomato leaves individual changes in life-history traits after 25 generations and then calculated growth rates from female individual measures; an additional one-generation transfer on tomato leaves before measures allowed control of potential genotype × environment effects. After 25 generations, growth rate of individuals originating from the cucumber ancestral populations was identical between the replicate lines maintained on cucumber and those transferred on tomato plants, despite the male adult survival being higher for the latter. In contrast, growth rate was higher for individuals transferred on tomato than those maintained on bean plants, mainly due to an increase in female fecundity and a decrease in the development time of both male and female individuals transferred on tomato plants. We are now comparing these data obtained on tomato leaves to similar data measured on entire tomato plants so as to evaluate potential differences in induced-defenses between leaves and plants, and subsequent potential differences in adaptation measures.

BIOINFORMATIC ANALYSIS OF AMPLICON SEQUENCING DATA TO STUDY SPATIAL AND TEMPORAL VARIATION IN A DAPHNIA MICROPARASITE

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Caullerya mesnili (Opisthokonta, Ichthyosporea) is an endoparasite infecting *Daphnia* (Crustacea, Cladocera) gut. This protozoan has high virulence and a strong genetic specificity for the infection, thus it seems to be a good model to study host-parasite coevolution dynamics. However, little is known about population structure of this microparasite. Previous work based on cloned sequences has shown that variation of the ITS region (internal transcribed spacer of ribosomal DNA) can be used to analyse spatial and temporal variation in *C. mesnili*. However, high-throughput next generation sequencing (NGS) allows much larger scale analyses. In this work we will present a bioinformatic pipeline analysing 392 bp long ITS amplicons from *Caullerya* obtained from 454 pyrosequencing. With this approach, it is possible to study in detail aspects such as the spatio-temporal distribution of *Caullerya* in different host populations or clones. We will also demonstrate the comparison of patterns obtained by cloning with the NGS approach.

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THE EVOLUTION OF CRISPR-CAS IN MYCOBACTERIUM TUBERCULOSIS

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The genomic regions encoding CRISPR-Cas (clustered regularly interspaced short palindromic repeats CRISPR associated proteins) are involved in generating adaptive immunity against invading genetic elements in many bacterial species. In Mycobacterium tuberculosis, the causative agent of human tuberculosis, the CRISPR-Cas region has a relatively high level of polymorphism in the number of repeats and spacer sequences. Yet, a relation between the CRISPR-Cas region in M. tuberculosis and immunity to foreign genetic elements has not been demonstrated. A search for an homologue region in other related mycobacteria revealed that the CRISPR-Cas region is only present in M. tuberculosis and in M. canettii. This suggests that the CRISPR-Cas region was acquired via horizontal gene transfer in the common ancestor of both species. In order to get some insight into the genetic variation of the CRISPR associated Cas proteins in M. tuberculosis, we analyzed deletions and SNPs in the Cas genes of 249 strains collected worldwide. The results indicate that Cas gene deletions are common in strains belonging to the East-Asian M. tuberculosis lineage, less common in other so-called modern lineages but completely absent from basal lineages. Contrastingly, substitution rates in Cas genes are similar among different phylogenetic lineages and mostly indicate strong purifying selection, except in Cas6 and Cas10. The CRISPR-Cas region in M. tuberculosis seems thus to be under selection to be preserved for unknown reasons. However, it is eroding in certain modern lineages indicating that its original function is either not preserved or is being compensated by other CRISP-Cas genes which were not uncovered in our analysis.

ANTLER QUALITY IN RED DEER: A TEST OF HAMILTON AND ZUK HYPOTHESIS

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The evolution and maintenance of elaborate secondary sexual traits in males has been the subject of intense interest since Darwin. Hamilton and Zuk (1982) hypothesis of parasite-mediated sexual selection suggests that genetically resistant males can afford to invest more in costly ornaments. Therefore elaborated sexual traits can serve as honest indicators of male health and parasite load. We study the association between the MHC (Major Histocompatibility Complex) class II genes, gastro-intestinal and lung parasite burden and the development of antler (sexual ornament) in red deer (*Cervus elaphus*). We analyzed associations between antler elaboration (mass and 8 other measurements) and parasite burden (lung nematode larva, abomasum nematodes and fecal egg counts). We found a very complex pattern of relationships. The mass of antler was significantly affected by the lung nematode larva burden. We used 8 antler measurements to describe antler size using principal component analysis (PCA). PC1 (explaining 80% of variance in antler size) was significantly affected by lung nematodes burden and PC3 (explaining 5% of variance) by abomasum nematode burden. We will additionally present results of analyses investigating the effect of MHC genotype on parasite load.

HOW MALARIA GETS AROUND: THE GENETIC STRUCTURE OF A PARASITE, VECTOR AND HOST COMPARED

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Parasites with complex life cycles have two host species shaping their genetic structure, but the traditional view posits that the parasite's structure will be mainly determined by the most mobile host species. Malaria causing protozoa are a prime example of parasites with a complex life cycle, needing both a dipteran and vertebrate host to complete their life cycle. In both hosts they impose selection pressures and are known to have reshaped host populations both demographically and behaviourally. Yet, how these host populations shape the parasite's genetics has been little studied. Especially the role of the dipteran vector has often been neglected. The relative contribution of each host to the parasite's population structure has therefore never been satisfyingly determined. In this talk we will compare the genetic structure of all three actors in a parasite-vector-host system: the vertebrate host, the longfingered bat (Miniopterus schreibersii), a long distance seasonal migrant; the vector/dipteran host, the wingless bat fly Nycteribia schmidlii; and the malaria parasite Polychromophilus melanipherus, a close relative of the malaria parasite *Plasmodium* spp. Being the most mobile host, our hypothesis was that M. schreibersii will show the most similarities with the structure of P. melanipherus. Traditional microsatellite markers, *cytb* DNA sequencing and a SNP library obtained by ddRAD-tagging reveal relatively high structure in the bats. In contrast, the haplotype distribution of *P. melanipherus* shows little geographic differentiation. Using both pattern-based and process-based coalescence methods we will try to demonstrate that the dipteran host plays a vital role in the observed patterns. In the end the specific life history traits of the vertebrate and dipteran host together are responsible for the found patterns and these are therefore discussed within the frame work of coevolution.

PRIVATE SELECTIVE SWEEPS IDENTIFIED FROM NEXT-GENERATION POOL-SEQUENCING REVEAL CONVERGENT PATHWAYS IN TWO INBRED SCHISTOSOMA MANSONI STRAINS

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The trematode flatworm of the genus Schistosoma, the causative agent of schistosomiasis, is among the most prevalent parasites in humans, affecting more than 200 million people worldwide. In this study, we focused on two well-characterized strains of S. mansoni, to explore signatures of selection underlying phenotypic variation. Both strains are highly inbred and exhibit differences in life history traits, in particular in their compatibility with the intermediate host Biomphalaria glabrata. We performed high throughput sequencing of DNA from pools of individuals of each strain using Illumina technology at an overall 20-fold genome-wide coverage. As a whole, 708,898 SNPs were identified and roughly 2,000 copy number variations. The SNPs revealed low nucleotide diversity ($\pi = 2 \times 10-4$) within each strain and a high differentiation level (Fst = 0.73) between them. Based on a recently developed in-silico approach, we further detected 12 and 19 private (i.e. specific non-overlapping) selective sweeps among the 121 and 151 found in total for each strain respectively. Interestingly, functional annotation of transcripts lying in those private selective sweeps revealed specific selection for functions related to parasitic lifestyle (e.g. cell-cell adhesion or oxydo-reduction). Despite high differentiation between strains, we identified an evolutionary convergence for proteolysis, known as key virulence factor and potential target for drug and vaccine development.

LAST MINUTE PASSENGERS OR PRIORITY TRAVELLERS: VIEW OF A WOLBACHIA JOURNEY IN ARMADILLIDIUM VULGARE OVARIES

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Wolbachia is a widespread endosymbiotic bacterium of arthropods and nematods. With the aim to better understand close interactions linking Wolbachia and its hosts, we study mechanisms of Wolbachia entry and long-term installation in a host easy to handle, the terrestrial isopod Armadillidium vulgare. Wolbachia is vertically transmitted to the host offspring, even though ovary cells are cyclically renewed. Thus, we search to identify mechanisms allowing Wolbachia to enter and to maintain itself in oocytes. Using Fluorescent in situ hybridization (FISH), we showed that the proportion of infected ooocytes increases in the course of ovary and oocyte maturation. At the end of ovary maturation, this proportion reaches the transmission rate to offspring. This enrichment can be explained by a secondary acquisition of the bacteria by oocytes (Wolbachia can be seen as last minute passenger) and/or by a preferential selection of oocytes infected with Wolbachia (as priority traveller). To confirm our first hypothesis, we highlighted Wolbachia's ability to infect oocytes along maturation by transinfection. Cellular mechanisms then need to allow Wolbachia internalization into the cell, without its destruction. Wolbachia also needs to intercept components required for its proliferation. To identify the processes involved, we monitored metabolic pathways via a host mRNA detection method (Single molecule FISH) compatible with our Wolbachia detection. The novelty of this approach is that we detect mRNA expression for the entire organ. Comparing uninfected ovaries, naturally infected ones and ovaries in the course of invasion after transinfection with Wolbachia, we denoted some different mRNA response patterns. For mRNA markers reporting the activation of internalization pathways, we observed an expression in all oocytes although Wolbachia was not present in each cell. In other cases, we could observe different mRNA sub-cellular localization, even some co-localizations with Wolbachia.

GENETIC/EPIGENETIC MECHANISMS TO TRIGGER OFF SEPARATE SEX APPEARANCE EXAMPLE OF INTRIGUING DIOECIOUS PARASITE: SCHISTOSOMATIDAE

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Sex determination and gonadal differentiation have always aroused a strong interest in evolutionary biology. Sexes appeared independently through all the animal kingdom. For instance, among the 18.000 species of classically hermaphroditic Trematodes, the hundred species of Schistosomatidae are intriguing because they possess separated sexes. In Schistosomes it has been proposed that the apparition of sexes resulted from host pressure when the hermaphroditic ancestors had colonized the warm-blooded hosts having a more sophisticated immune system. Sex of Schistosomes is genetically determined in the zygote at fertilization, but there is no phenotypic dimorphism in all larval stages: sexual dimorphism only appears in adult worms, in the vertebrate definitive host. From a cytogenetic point of view, males are homogametic ZZ and females are heterogametic ZW. The W heterochromosome presents a unique feature: it is devoid of W specific genes but instead, it presents satellite repeat sequences. Moreover, the chromatin structure of these satellite repeat sequences changes through the different stages of parasite life cycle. These elements argue in favor of an original epigenetic mechanisms triggering for sex appearance in Schistosomes. But this does not exclude any possible genetic determinant. These uncommon ZW Trematodes display four putative testis associated genes: Doublesex/mab-3 related (Dmrt) family of transcription factors. In order to connect the apparition of phenotypic sexual dimorphism to molecular mechanisms we have studied the expression on these Dmrt candidates on Schistosomula, the stages where the differentiation appears. Besides, we carried out whole genome approaches: RNA and ChIP (Chromatin ImmunoPrecipitation) sequencing on the same developmental stages. These powerful new technologies and the coming of new epigenetic concepts should offer a new gaze on these important, yet unsolved old evolutionary questions.

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RECENT RAPID SPREAD OF WOLBACHIA VARIANTS IN EAST AUSTRALIAN DROSOPHILA SIMULANS: INDUCED BY PROTECTION AGAINST NATURALLY OCCURRING PATHOGENS?

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The maternally inherited intracellular bacteria Wolbachia are ubiquitous amongst arthropod hosts. Although well known for their ability to induce various host reproductive manipulations, Wolbachia have also been shown to increase host fitness by protecting against infectious microbes or increasing fecundity; and they are expected to evolve towards mutualism in natural host populations. Recent data from our lab indicates the rapid sequential spread of two Wolbachia variants (wAu and wRi), only one of which induces significant reproductive parasitism in the form of cytoplasmic incompatibility, in Australian populations of Drosophila simulans over approximately 20 years. In each case analyses suggest that these dynamics are best understood as Fisherian waves of favourable variants, involving net host fitness benefits of a non-trivial magnitude. Despite their leading roles in genetics, the suite of naturally occurring pathogens which significantly impact field populations of *Drosophila melanogaster* and D. simulans is not well characterized. Moreover specific infections (such as DMelSV) that are maintained for study in laboratory populations may have relatively benign effects in nature. We have found that each of the wAu and wRi Wolbachia variants currently persist, at quite different infection frequencies, amongst geographically isolated populations of *D. simulans* within Australia. If these Wolbachia variants have the effect of promoting host fitness by protecting against infectious microbes, a comparison of these different host populations has potential to reveal pathogens that are ecologically significance and provide some indication of the intensity of the selective pressure they exert on a genetically tractable model host.

CELL SACRIFICE IN THE GUT: AN ADAPTIVE RESPONSE TOWARDS NOSEMA SPP. INFECTION IN HONEY BEES

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The microsporidian *Nosema ceranae* is a natural parasite of the Asian honeybee *Apis cerana* but is now also a widespread cause of Nosemosis in the European honey bee *A. mellifera*. *N. cerana* infections can have severe effects on honey bee fitness at the individual and colony level. We found that the extensive breeding effort by Danish beekeepers against the native microsporidian parasite *N. apis* has produced a *Nosema* tolerant honey bee strain, which revealed a strongly up-regulated immune response when challenged by *N. cerana* infection compared to an unselected strain. After transmission via the fecaloral route, spores normally germinate in the midgut, where they penetrate, replicate and destroy the cells of the gut epithelium. To understand the effect of the altered immune response on the level of infestation and destruction of the midgut epithelium, we compare sections between the selected and an unselected strain over the course of infection. We discuss these results in the context of an adaptive immune response and other underlying biological mechanisms of the selected strain against *N. ceranae* infection.

TESTS OF COEVOLUTION BETWEEN VIRUS AND HOST IN KILLER YEAST STRAINS

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Viruses are among the most abundant biological entities on earth and found in almost every organism. One of their strategies is to adapt to their host and develop a mutualistic endosymbiosis. In case of the "yeast-virus killer system", the cytoplasmic virus produces a compound that is toxic to yeast strains without the virus, while the yeast cell allows for viral survival and reproduction. Transfer of the virus is strictly vertical. The benefit of competitor killing for the host is thought to depend on several factors, including the phylogenetic relatedness between killer and sensitive strain and the structure of the environment. However, presence of viruses was described for a limited number of laboratory strains, and little is known about their frequency and ecological importance in nature. Among a worldwide collection of 184 yeast strains of known phylogenetic history, we identified 14 strains infected by killer viruses. We classified and assigned them into one of three types: K1, K2 or K28, where the most common appeared to be K1. We test for the signature of coevolution between these killer viruses and their yeast hosts in two ways. First, we test the killing ability of viral carriage as function of host phylogenetic distance, including self versus non-self to see if they are found least toxic towards their own hosts as a result of the co-evolution between virus and host. This is done by cross-infecting strains with the viruses they contain, combined with analyses of their toxicity. Second, we will compare the phylogenies of virus and host based on their genome sequence to learn whether the viruses tend to move freely between different hosts.

THE EVOLUTION OF RECENT ANTIVIRAL GENE DUPLICATES IN DROSOPHILA

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RNAi is a major invertebrate defensive pathway, in which small RNAs are derived from a target RNA and guide an Argonaute-family protein to cleave and subsequently degrade the target. This mechanism defends against both viruses and transposable elements. These contrasting and potentially conflicting selective pressures may have driven the rapid adaptive evolution documented in Argonaute-family genes, which places them among the top 3% of the fastest evolving D. melanogaster genes. It has been hypothesised that this rapid evolution is driven by an arms race between viral suppressors of RNAi (VSRs) and the RNAi mechanism. In Drosophila, Argonaute2 (Ago2) is the effector protein in siRNAmediated antiviral and anti-TE defence. The majority of insects possess a single Ago2; however, duplication has occurred in several Dipteran taxa including Phoridae, Glossina and Drosophila. Ago2 has undergone numerous recent duplications in the obscura group of Drosophila, producing two paralogues in D. subobscura, three in D. obscura and five in D. pseudoobscura. We present population genetic data for the Ago2 paralogues in D. subobscura, D. obscura and D. pseudoobscura. We find that most paralogues have remarkably low genetic diversity, possibly resulting from recent selective sweeps. We also find strong evidence for codon usage bias in the earliest-diverging paralogue. Additionally, we present expression data in different tissues and in response to viral exposure. These results suggest that the Ago2 paralogues in the obscura group have each evolved under different selection pressures, possibly imposed by functional specialization in an evolutionary arms race.

HOST ADAPTATION IS CONTINGENT UPON THE INFECTION ROUTE TAKEN BY PATHOGENS

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Horizontally-transmitted pathogens can infect their hosts through different routes. Yet, the physiological and evolutionary consequences to the host of distinct modes of pathogen access are virtually unknown. To tackle this question, we used Experimental Evolution of Drosophila melanogaster infected with Pseudomonas entomophila by two different routes (oral and systemic). We found that adaptation to both routes relied on resistance. Moreover, adaptation to infection through one route did not protect from infection through the alternate route, indicating distinct genetic bases. Also, the two selection regimes led to markedly different evolutionary trajectories. Finally, relatively to the control population, evolved flies were not more resistant to bacteria other than Pseudomonas and showed higher susceptibility to viral infections. These specificities and trade-offs may contribute to the maintenance of genetic variation for resistance in natural populations. Moreover, our data shows that pathogen infection route affects host evolution. Therefore, the study of host-pathogen interactions should account not only on host and pathogen evolution, but also on the ecology of the infection, when interpreting patterns of variation in natural populations.

REMARKABLE DIVERSITY OF ENDOGENOUS VIRUSES IN THE GENOME OF AN ISOPOD CRUSTACEAN

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Recent studies in paleovirology have uncovered myriads of viral genome fragments integrated in the genome of their eukaryotic hosts. These fragments result from endogenization, i.e., integration (often accidental) of the viral genome into the host germline genome followed by vertical inheritance. So far, most studies have used a virus-centered approach, whereby endogenous copies of a particular group of viruses were searched in all available sequenced genomes. Here we follow a host-centered approach whereby the genome of a given species (the crustacean isopod Armadillidium vulgare) is screened for the presence of endogenous viruses using all viral genomes sequenced to date (n = 2048) as queries. This search and downstream evolutionary analyses revealed that 50 viral genome fragments belonging to 10 viral families became endogenized in A. vulgare. We show that viral endogenization occurred recurrently during the evolution of isopods and that some endogenous virus loci are polymorphic in A. vulgare, suggesting they result from recent endogenization of viruses likely to be currently infecting isopod populations. Overall, our work shows that isopods have been and are still infected by a large variety of DNA and RNA viruses. It also extends the host range of several families of viruses and brings new insights into their evolution. More generally, our results underline the power of paleovirology in characterizing the viral diversity currently infecting eukaryotic taxa.

SEX-SPECIFIC GENETIC VARIANCE FOR RESISTANCE AND TOLERANCE TO INFECTION

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Understanding why individuals express a given level of immune competence is necessary for predicting the incidence, spread, and evolution of disease. It is often observed that males have weaker immune systems than females and thus get sick more often. Sex-biases in susceptibility, both before and during infection, can affect pathogen prevalence and host-parasite coevolution. One potential cause of sexual dimorphism in immunity could be differential effects of infection on fitness between the sexes, i.e. females may be selected to invest more in immunity because the fitness costs of infection are greater. When individuals differ in their ability to maintain fitness during infection they are said to express different levels of tolerance. Most eco-immunology studies assess resistance, which involves those responses that prevent, restrict, or clear infection by parasites, rather than tolerance mechanisms. Distinguishing between resistance and tolerance is important as they predict markedly different evolutionary and epidemiological outcomes. Tolerance has long been acknowledged as a mechanism of managing stressors in the plant literature, but few studies have investigated tolerance in animals. To date, no comparison of tolerance in male and female animals has been undertaken. Using males and females from several Drosophila melanogaster genotypes, we assessed resistance and tolerance to the common bacterial pathogen Pseudomonas aeruginosa. Across all lines, resistance and tolerance was significantly lower in females. There was a significant negative correlation between male and female tolerance. In addition, we observed a genetic trade-off between resistance and tolerance. Our results suggest that antagonistic pleiotropy can maintain genetic variation in immunity. Implications for the genetics of immune function and sex-specific immunocompetence will be discussed.

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WALKING WITH INSECTS: MOLECULAR MECHANISMS BEHIND PARASITIC MANIPULATION OF CATERPILLAR BEHAVIOUR

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Parasitic modification of host behaviour is a widely adopted strategy of parasites to enhance their own transmission. The examples of behavioural manipulation are rapidly accumulating, covering a broad spectrum of parasites and hosts. Nevertheless, surprisingly little is known on the underlying causative molecular mechanisms. A typical case of behavioural manipulation is found in insects infected with baculoviruses. Infected caterpillars show enhanced mobility and start climbing to the top of plants or the forest canopy ('tree top disease'). As a consequence, the virus is spread over a larger area, thereby increasing the chance to infect a new caterpillar. The baculovirus-insect system provides an excellent platform to study parasitic manipulation of insect host behaviour. It allows the comparative analysis between wildtype viruses and single gene knock-out mutants. Recently, the *eqt* gene from the baculovirus Lymantria dispar nucleopolyhedrovirus (LdMNPV) was identified to induce tree top disease in *Lymantria dispar* larvae. Here, we studied the effect of *Autographa californica* multiple nucleopolyhedrovirus (AcMNPV) on climbing behaviour in two different host insects, the cabbage looper *Trichoplusia ni* and the beet armyworm *Spodoptera exiqua*. We show that the effect of this virus on caterpillar behaviour differs between these two host species. Additionally, we found no evidence for a general role of the *eqt* gene in causing tree top disease. This implies that baculoviruses have evolved multiple strategies to manipulate similar behavioural phenotypes in their caterpillar hosts.

GENETIC STRUCTURE OF A NEMATODE PARASITE (TRICHURIS MURIS) ACROSS THE EUROPEAN HOUSE MOUSE HYBRID ZONE

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A continuous arms race between hosts and parasites may lead to co-adaptations varying both spatially and temporally. In secondary contact hybrid zones between divergent host taxa, these co-adaptations may break down, leading to a higher/lower prevalence of parasites in hybrids and a barrier to parasite gene flow at the centre of host contact. Here we aimed to study the genetic structure of a common gastrointestinal nematode parasite, Trichuris muris (Trichuridae) of the house mouse (Mus musculus) in the European house mouse hybrid zone (HMHZ). Specifically, we asked i) Are parasites genetically structured according to the host hybrid zone? ii) Could there also be a hybrid zone between the parasites? We analyzed 172 worms from 47 localities (one nematode per wild caught house mouse) covering an area of 6,500 km2 in Central Europe (Czech Republic and Germany). We sequenced the mitochondrial cytochrome oxidase-I (COX-I) gene and genotyped three microsatellite markers from these parasites. We found 22 haplotypes in COX-I sequences which, by performing haplotype network analysis grouped into two large and two small clades. However, the geographic pattern of these parasite mt clades did not correlate with host genotype across HMHZ. Hierarchically clustering (Structure, k=2) the parasite microsatellite data suggests two clusters, but again with no correlation to the east-west host contact. Our results strongly contrast with the genetic structure of another nematode, Syphacia obvelata which mirrors the host HZ geography. We suggest this contrast is due to the lesser host specificity of T. muris compared to S. obvelata. In the sampled field area, the house mouse is sympatric with woodmice (genus Apodemus) or rats (Rattus norvegicus), both being common hosts of T. muris. Such alternative hosts provide a way round any transmission barrier. In conclusion, T. muris is not genetically structured according to its host in the HMHZ and there is no evidence of hybrid individuals.

9. Genetics and Genomics of Host-Parasite Coevolution

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HOST SPECIES SPECIFICITY OF BORRELIA AFZELII STRAINS

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Most pathogens consist of a number of antigenically different strains. The coexistence of several antigenic variants is often considered to be a result of diversifying selection from host acquired immunity. In multi-host pathogens (i.e., pathogens that infect >1 host species), an additional potential cause of antigenic diversity is that different strains are adapted to different host species. Here, we tested for such host species specificity of strains of the tick-transmitted bacterium *Borrelia afzelii*. The main hosts of *B. afzelii* are rodents and shrews. *B. afzelii* can be divided into strains based on outer surface protein C genotype (*ospC*) and individual hosts are often infected by multiple *ospC* types. We used 454 amplicon sequencing to resolve the strain composition of infections in bank voles, yellow-necked mice and common shrews. Among 123 infected individuals, we found seven different strains. All strains were found in all host species, but the prevalence of strains varied in a species-specific manner. Thus, host specificity may contribute to the maintenance of antigenic diversity in this pathogen.

THE POTENTIAL FOR GENE-FOR-GENE AND MATCHING-ALLELE COEVOLUTION IN A PROTIST HOST-PARASITE SYSTEM

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The dynamics and consequences of host-parasite coevolution depend on the nature of host genotypeby-parasite genotype interactions (G×G) and whether there are costs of resistance and infectivity. Gene-for-gene interactions (GFG) without costs result in selective sweeps with generally low levels of genetic variation. In contrast, matching allele interactions (MA), and GFG with costs, easily give cyclic dynamics of allele frequencies and high levels of genetic variation. Here we investigate the relative potential for GFG and MA coevolution in a protist host-parasite system, the dinoflagellate *Alexandrium minutum* and its parasite *Parvilucifera sinerae*. We challenged 9 different isolates of *A. minutum* with 10 isolates of P. sinerae in a fully factorial design and measured infectivity and host and parasite fitness. There were strong G×Gs for all traits. About 2/3 of the G×G variance components were due to MA type interactions. There were no costs of resistance or infectivity. We conclude that here is high potential for cyclic dynamics as a result of MA interactions, but also some scope for selective sweeps as a result of GFG without costs.

9. Genetics and Genomics of Host-Parasite Coevolution

D21SY09PS0928

CHARACTERIZATION OF CANDIDATE GENES FROM A QTL ANALYSIS OF THE SKIN MICROBIOTA IN HOUSE MICE

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Meriem BELHEOUANE1,2, Saleh IBRAHIM3 and John F. BAINES1,2 1Institute for Experimental Medicine, Christian-Albrechts-University of Kiel, Germany 2Max Planck Institute for Evolutionary Biology, Plön, Germany 3Department of Dermatology, University of Lübeck, Lübeck, Germany The skin is a complex ecosystem inhabited by diverse microbial communities, and various factors including host genetics, immune status and the environment influence community structure and diversity over space and time. Several skin diseases are postulated to have a microbial component, but little is known about the underlying mechanisms or origins of disease susceptibility. To measure the host genetic contribution to the structure and diversity of the skin microbiota and its potential contribution to disease, we performed quantitative trait locus (QTL) mapping of both autoimmune skin blistering and bacterial traits in an advanced intercross between house mouse strains derived from multiple subspecies. To understand the evolutionary origin of host genetic variability influencing both individual bacterial abundances and susceptibility to disease, we are subjecting candidate regions to more detailed molecular population genetic analysis in natural populations of house mice. In parallel, bacterial species with putative probiotic effects will be cultured and subject to genomic analysis to shed light on the role of host - commensal microbe coevolution in maintaining homeostasis of the skin community.

ERYTHROCYTE POLYMORPHISMS ASSOCIATED WITH PROTECTION AGAINST P. VIVAX MALARIA IN PAPUA NEW GUINEA CHILDREN

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Malaria parasites have long coexisted with hominid ancestors: P. falciparum might had infected humans before the out-of-Africa migration and followed the human expansion throughout the tropics. This long-term interaction may have driven the co-evolution of the host-parasite and left evolutionary foot-prints in both human and parasite genomes. According to the "malaria hypothesis", the high frequencies of deleterious mutations of some human populations are probably due to the strong selective pressure of malaria A remarkable range of polymorphisms have been associated with protection against malaria. Populations of the South West Pacific, a co-endemic region for all four human malaria parasite species, are highly diverse and exhibit a range of unique red blood cell polymorphisms, with geographical patterns paralleling malaria endemicity. We conducted three independent studies in the Madang area of Papua New Guinea to analyse the association of South East Asian Ovalocytosis (SAO) (caused by band 3 deletion SLC4A1Δ27) and Gerbich negative blood group (caused by a deletion in exon3 of the glycophorin C gene (GYPC)), with protection against P. vivax malaria, testing the hypothesis that P. vivax malaria contributed to the selection of those polymorphisms. Our results showed that SAO was associated with a 46% reduction in risk of clinical P. vivax episodes in a cohort of infants 3-21 months and a 52-55% reduction in P. vivax reinfection in children 5-14 yrs. A case-control study of children 0.5–14 yrs revealed the potential for a strong protection against severe P. vivax malaria. Analysis of Gerbich blood group variants in a cohort of children 1-3 yrs showed that GYPC homozygote is associated with strong protection against P. vivax malaria and that the strength of protection increases with parasitemia. This strongly suggest that P. vivax malaria may have contributed to shaping the unique host genetic adaptations to malaria in Asian and Pacific populations.

TRACING THE EVOLUTIONARY HISTORY OF KOALA RETROVIRUS: FREQUENT HOST-SHIFTS IN A VIRUS LINEAGE OF DISEASE AND CONSERVATION RELEVANCE

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Retroviruses are a diverse family of viruses with a unique replication mechanism involving reverse transcription and integration into the host genome. Occasionally, a retrovirus may integrate into a host germ-line cell, leading to vertical transmission as an endogenous retrovirus (ERV). Consequently, vertebrate genomes contain numerous ERVs of varying ages (eg ~8% of the human genome), acquired during their ancient co-evolutionary relationship with retroviruses. Active exogenous retroviruses are causative agents of several serious diseases, and in some cases may undergo considerable taxonomic shifts to infect new host species. An example is koala retrovirus (KoRV), which is associated with the development of tumours and chlamydiosis in hosts. KoRV is believed to have entered the koala population within the last 100 years, with levels of infection reaching 100% in northern Queensland and only one population in Australia remaining uninfected. The origin of koala retrovirus is currently unclear, but it shares similarity with retroviral sequences isolated from gibbon apes, pigs and mice. In this study 90,000 high quality ERV sequences from 60 vertebrate genomes were analysed to investigate the evolutionary history of the KoRV lineage. Extraction of useful phylogenetic signal from retroviral sequences is complicated due to their small genome size and rapid evolution. Consequently a novel phylogenetic method for extracting informative evolutionary signal from retroviral sequences was implemented. The results reveal a history of extensive host shifts across vertebrate diversity, and provide clues regarding the potential origin and spread of KoRV. The findings also demonstrate the vast uncharacterised diversity of ERV sequences in vertebrate genomes, which present a valuable resource for investigating host-parasite coevolutionary patterns over time.

THE SPIRONUCLEUS SALMONICIDA GENOME

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Eukaryotic microbes are highly diverse and remain understudied. Among them, diplomonads is a group of binucleate heterotrophic flagellates. The most studied diplomonad is *Giardia intestinalis*, an intestinal parasite in humans and other animals causing diarrhea. Spironucleus salmonicida, the focus of this study, causes systemic infections in salmonid fish. We have developed a stable transfection system for S. salmonicida and identified energy-producing hydrogenosomes, rather than mitosomes like in *Giardia*. Here we present the analysis of the whole genome sequence. The genome of S. salmonicida is ~12.9 MB, similar to the size of the Giardia genome. There are 8,034 protein coding genes with ~2500 more than in Giardia. The two genomes share similar machineries for intron splicing, DNA replication, transcription, RNA processing, and most metabolic pathways. However, there are several clear differences. S. salmonicida shows signs of more regulations on the transcription level with promotor-like motifs identified for >1,000 highly expressed genes. It utilizes an alternative genetic code in which the sole stop codon also serves as the core motif in the polyadenylation signal. S. salmonicida like Giardia has a large family of cysteine-rich variant-surface proteins but lacks the RNA interference pathway which is involved in their regulation. Otherwise, *S. salmonicida* has an extended metabolism compared to *Giardia*. For example, there is an expanded repertoire of genes involved in oxidative stress response. The oxygen level within the fish fluctuates and efficient scavenging of reactive oxygen species is needed to avoid damage to the sensitive hydrogenosomal enzymes. Many of these genes have been acquired from anaerobic bacteria, probably as an adaptation to a pathogenic lifestyle. The S. salmonicida genome will not only be valuable for the research on diplomonads, but also for studies of the diversity of eukaryotic processes.

SEQUENTIALLY VARYING REPRODUCTIVE STRATEGIES IN A PARASITIC FLATWORM: RIDING A POPULATION GENOMIC ROLLER COASTER

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Breeding systems have major impacts on population genetic processes. At ecological time scale, asexuality is a superior reproductive strategy, because it provides the ability to colonize new environments quickly and allows populations to grow exponentially in favorable conditions. In spite of high fitness in short term, small effective population size and lack of effective recombination reduce the adaptive potential and eventually lead to accumulation of deleterious mutations. Thus, at evolutionary time scale, sexual reproduction becomes more advantageous. Our study focuses on the genus *Gyrodactylus* (Platyhelminthes, Monogenea), which consists of small host specific fish parasites. In this genus, combination of clonality and sexuality has proved to be an ideal strategy to be successful at both time scales. New genetic combinations are created by occasional sexual reproduction, and the fittest genotypes can then be amplified by clonal parthenogenesis and spread to new geographical areas, assuming a suitable vector like farmed salmonids. Over time, rare sexuality in a population consisting of few genotypes and increasing selection pressure by the host cause the gene pool to become even narrower, and the declining fitness leads inevitably towards extinction. However, the genus is extremely species-rich, containing perhaps thousands of specific lineages on different hosts. The rescue seems to happen through host switching by hybridization among distant lineages. These switches provide a way out of the otherwise inescapable evolutionary dead-end, and bring the riders back to the highest peak of the population genomic roller coaster.

REPRODUCTIVE BARRIERS AND CANDIDATE GENES INVOLVED IN THE ISOLATION OF FOUR CLOSELY RELATED SEXUALLY DECEPTIVE ORCHIDS

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Sexually deceptive orchids of the genus *Ophrys* have a highly specialized pollination system in which each orchid species is thought to attract only one or very few specific pollinator species. This study aims at dissecting the ecological and molecular mechanisms of reproductive isolation between four closely related and sympatric *Ophrys* species (*O. exaltata, O. sphegodes, O. garganica* and *O. incubacea*) that specifically attract males of four different pollinator bee species. Flowering phenology and pollinator behaviour were found to act as pre-mating reproductive barriers among these species, but crossing experiments and ploidy analysis revealed no post-mating barriers. Transcriptome sequencing allowed the identification of candidate genes involved in specific pollinator attraction and reproductive isolation. This approach identified candidate genes for hydrocarbon (odour) and anthocyanin (colour) biosynthesis, as well as transcription factors putatively involved in the regulation of flower odour, colour and morphology.

9. Genetics and Genomics of Host-Parasite Coevolution

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HOST DISPERSAL SHAPES HOST-PARASITE CO-EVOLUTIONARY DYNAMICS IN CICHLID FISHES

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Parasite communities have been proposed to drive adaptation and reproductive isolation of their hosts mediated by the pleiotropic role of immunogenes in both parasite defence and assortative mating. Parasite communities themselves are shaped by the abiotic environment, the availability of intermediate hosts, and live history traits of the host species. The degree to which parasites drive the evolution of their hosts hence may directly rely on intrinsic host traits. Here, we tested the hypothesis whether host dispersal can drive parasite community structure and investigated how this influences the degree local immunogenetic adaptation of the host. We compared parasite communities and Mhc (major histocompatibility complex) allele frequencies in two sympatric cichlid fish species with contrasting dispersal patterns along the shoreline of Lake Tanganyika. We found that allopatric and genetically diverged colour morphs of philopatric blunt-headed cichlids (*Tropheus* spp.) are infected by contrasting parasite communities and are immunogenetically adapted to them. We could not detect such a pattern in the closely related, but non-philopatric cichlid fish species *Simochromis diagramma*. This suggests that the presence of migrants among isolated host populations homogenises their respective parasite communities in Lake Tanganyika and ultimately affects immunogenetic adaptation. Therefore we conclude that host-parasite co-evolutionary dynamics are the main driver of diverging parasite communities among allopatric host populations. The proposed pleiotropic role of the *Mhc* in parasite defence and assortative mating lets us speculate, whether contrasting parasite communities could drive speciation in colour morphs of philopatric blunt-headed cichlids.

THE GENETIC BASIS OF VARIATION IN ANTIVIRAL DEFENSE IN DROSOPHILA MELANOGASTER

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The presence of evolutionary effects that pathogens and hosts have on one another provides an excellent model to study co-evolution. To understand the molecular basis of how insects evolve resistance to viruses in nature, we have investigated the genes that cause variation in the susceptibility of *Drosophila melanogaster* to viral infection. Two viruses that naturally infect *D.melanogaster*: sigma virus (DMelSV) and Drosophila C virus (DCV) have been used in this study. DMelSV infects up to 18% of flies in natural populations and is therefore naturally coevolving with flies. It is only transmitted vertically from parent to offspring and is a host-specific pathogen of *D.melanogaster*. In contrast, DCV infects many Drosophila species and can be transmitted horizontally. By using recombination mapping and GWAS, we identified genes and polymorphisms in the genes that associated with variation in the susceptibility of *D.melanogaster* to DMelSV and DCV separately. In order to experimentally verify the roles of these genes, we carried out RNAi experiments to knock down target genes and analysed the changes of susceptibility in flies. We also generated transgenic flies carrying specific polymorphisms of interest and analysed their effects on viral resistance in *D.melanogaster*.

CONTRASTING EVOLUTIONARY PATTERNS IN RECENT AND DEEP TIME SUGGEST OLD ORIGINS FOR EAST-AFRICAN ARENAVIRUSES AND STRONG SPECIFICITY TO THEIR MULTIMAMMATE MOUSE HOSTS

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To understand the relationship between evolutionary histories of zoonotic RNA viruses and their hosts, we study patterns of viral divergence across a host geographic range structured by both recent and ancient events. We focus on East African arenaviruses closely related to the virus causing human Lassa fever in West Africa and which infect the same natural host, the multimammate mouse Mastomys natalensis. We sampled at regular intervals along a 200 km transect in Tanzania, capturing a total of 1284 multimammate mice, of which 57 harbored arenaviruses. Multilocus microsatellite, mitochondrial and Y chromosome markers reveal a cryptic host contact zone between two M. natalensis taxa along the transect, with one admixed central locality and the surrounding 11 localities dominated by one of either taxon. The host mitochondrial lineages split about 1 million years ago and the current zone is likely the outcome of secondary contact of *M. natalensis* geographic isolates some thousands of years ago, driven by Holocene climate oscillations. We show that each *M. natalensis* taxon hosts a very distinct arenavirus, yet both arenavirus species are present in the host-admixed locality. Both hosts and viruses show further genetic differentiation within each host-taxon's ranges, but on this level the geographic patterns of host and virus structure are not correlated. These observations suggest that recent host evolutionary history has little effect on recent arenaviral divergence (despite similar differentiation time scales), yet host-virus associations at a deeper taxonomic level have likely persisted since their shared isolation during the last glacial cycle, predating current ancestry estimates for these RNA viruses. Furthermore, if arenavirus species are unable to cross between pairs of taxonomically cryptic host sister taxa in geographic contact, a rapid arenavirus spread across M. natalensis' pan-African range, likely home to many such cryptic taxa, seems remote.

HOST SPECIFICITY IN A VECTOR BORN DISEASE

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Understanding the processes that generate genetic diversity and which ultimately lead to speciation is a long standing issue in evolutionary ecology. As originally proposed by JBS Haldane, host-parasite interactions play a crucial role in this context. Both host and parasite can exert a strong selective force and induce diversification. Host specificity of *Borrelia*, the agent of the most common vector borne disease in temperate regions, is suggested since 15 years. While different processes have been proposed, the mechanisms leading to host specificity and the high genetic diversity in *Borrelia* are still unknown. It is generally assumed that the genetic diversity of *Borrelia* cannot be attributed to the vector since they are mainly transmitted by a single generalist tick species - *Ixodes ricinus*. However host specificity of vectors has only rarely been tested and recent work suggests that it may be more frequent than previously thought. Here we investigate the degree of specialisation of *I. ricinus* by comparing the genetic variation of larval ticks on two important host species for Borrelia: the yellownecked mouse and the bank vole. After correcting for either within host individual structure or geographical structure, we find consistent support for a low but significant genetic differentiation between ticks harboured by the two sympatric host species. Our results suggest that the tick-vector may play a crucial role in the host specificity and the genetic diversity of *Borrelia*. Further studies on the genetic diversity of *Borrelia* as well as on vector competence in each tick population are now needed to estimate the potential contribution of the vector in the evolution of a common vector born disease.

9. Genetics and Genomics of Host-Parasite Coevolution

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CAN DIFFERENTIAL GENE EXPRESSION EXPLAIN DIFFERENTIAL HOST RESISTANCE?

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Our ability to find signatures of selection in the genome is rapidly increasing. As a consequence we now know that strong selection leading to local adaptation can affect many regions in the genome. However, not all local adaptations result in structural gene changes. Changes in regulation of existing genes might be at least as important. To what extend this happens and whether selection only affects genes directly related to the focal trait, or also other gene classes is for many cases unknown. Hostparasite systems are very suitable for testing the importance of gene expression alterations in local adaptation. Selection is often strong due to the fact that parasites can have a big impact of fitness of the individual, making it probable we can pick up alterations in expression. Start and finish of changes in gene expression are much easier to predict than for many other (somatic) traits and temporal an spatial fluctuations in parasite abundance are common in nature, which may favor changes in gene expression rather than genetic changes as they are much easier to reverse. We used the F2 generation of two populations of stickleback fish that differ in host resistance, in order to identify which genes are involved in resistance and to understand whether heritable differences in resistance across populations are linked to differential gene expression. We infected naive fish with Gyrodactylus gasterostei, a parasitic flatworm and tracked the infection by counting parasites on the skin over time. Targeting the beginning and peak of the infection, we used microarrays to identify differential gene expression across the genome. We will present whether differences in host resistance result in differential expression patterns among populations and whether infection only affects immune related genes or a wider set of genes, thereby contributing to a better understanding of the role of gene expression alterations in host adaptation to parasites.

9. Genetics and Genomics of Host-Parasite Coevolution

D21SY09PS1241

PHYLOGENOMICS: AN EVOLUTIONARY FRAMEWORK BRIDGING SCHISTOSOME GENOMICS AND BIOLOGY

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Phylogenomics has opened new frontiers in Science by connecting genomic diversity to evolutionary systems biology. Many studies have applied this approach to improve functional annotation of genes/proteins and to reconstruct phylogenetic relationships across different domains of life. Moreover, this evolutionary framework has a broader impact that is to bridge parasite genomics and biology with potential applications in biomedicine and biotechnology. Here we focus on the phylogenomics of Schistosoma (Platyhelminthes), which causes human schistosomiasis, a neglected disease affecting over 230 million people worldwide. By applying phylogenomics to the predicted proteome analysis of Schistosoma mansoni in comparison to other metazoans, we have shed light on the evolutionary processes that shaped host-parasite interactions over evolutionary time. Our work has focused on Schistosoma distinct protein families that play key biological functions, e.g. eukaryotic protein kinases (ePKs). Our findings indicated that each protein kinase group, family, and subfamily have a unique evolutionary history. Furthermore, the main processes shaping the evolution of S. mansoni ePKs include exon shuffling and gene duplication followed by divergence. We also identified S. mansoni mitogen-activated protein kinases that play pivotal roles in multiple cellular processes as described in other metazoans. Based on these findings, S. mansoni proteins of the MAPK pathway were chosen for knockdown by RNA interference assays. Our results showed that SmJNK (Smp_172240) has an important role in the parasite transformation and survival representing a potential drug target to disrupt the schistosome life cycle preventing disease progression. Altogether, this study provides an evolutionary view of the S. mansoni ePKs improving its functional annotation and allowing for a better assessment of the parasite genome complexity and lineage-specific adaptations potentially related to the parasitic lifestyle.

HERITABLE VARIATION IN PARASITE TOLERANCE AND RESISTANCE INFERRED FROM A WILD HOST-PARASITE SYSTEM

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As defense mechanisms against parasites, hosts have evolved two distinct strategies: resistance (i.e. prevent infection or limit parasite growth) and tolerance (i.e. alleviate the fitness consequences of infection).Distinguishing between these strategies has important evolutionary implications to predict the coevolutionary outcome of host-parasite interactions. Most theoretical models predict the maintenance of genetic variation in resistance but the fixation of tolerance along the course of hostparasite coevolution. Moreover, a negative genetic correlation is expected between these two traits. However, these predictions have rarely been characterized in wild host populations, and few are known about the environmental and genetic bases of tolerance and resistance. Here, we estimate quantitative genetics parameters for resistance and tolerance in Leuciscus burdigalensis, a freshwater fish species being parasitized by the fin ectoparasite Tracheliastes polycolpus. Using a long-term survey, we reconstructed a full-sib pedigree in our host population using microsatellites data. We confronted empirical pedigree to simulated pedigrees to estimate the reliability of molecular-based pedigrees.We then used Bayesian animal models to estimate (i) both genetic and environmental variation in resistance and tolerance respectively, and (ii) the genetic correlation between these two traits. The reconstructed pedigree included 670 individuals assigned in 302 full-sib families. Although simulations revealed that our pedigree was perfectible, we found it was powerful enough to unravel significant broad-sense heritability for both host resistance and tolerance (i.e. ~10-20% for both traits). Animal models also revealed environmental effects for both traits (i.e. > 35 %). We failed to detect significant genetic and phenotypic correlations between these two defense strategies.

CAPTURING THE POPULATION STRUCTURE OF MICROPARASITES: USING ITS-SEQUENCE DATA AND A POOLED DNA APPROACH

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The internal transcribed spacer (ITS) region of nuclear ribosomal DNA is a central target not only for molecular identification of different taxa and strains but also for analyses of population structure of wild microparasite communities. Importantly, the multi-copy nature of this region allows for successful amplification of low quantity samples of the target DNA, a common problem in studies on unicellular, unculturable microparasites. We analysed ITS-sequences from the protozoan parasite Caullerya mesnili (class Ichthyosporea) infecting waterflea (Daphnia) hosts, across several host population samples. We showed that analysing representative ITS-types (as identified by statistical parsimony networks) is a suitable method to address relevant polymorphism. The spatial patterns were consistent regardless of whether parasite DNA was extracted from individual hosts or pooled host samples. Remarkably, the efficiency to detect different sequence types was even higher after sample pooling. As shown by simulations, an easily manageable number of sequences from pooled DNA samples was already sufficient to resolve the spatial population structure in this system. In summary, the ITS-region analysed from pooled DNA samples can provide valuable insights into the spatial and temporal dynamics of microparasites. Moreover, for the analysis of sequence variation in multi-copy gene regions, the application of statistical parsimony network analysis is clearly advantageous.

LINKING PARASITE GENETICS WITH HOST DISEASE PHENOTYPE, THE CASE FOR HUMAN SCHISTOSOMIASIS

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Schistosomiasis is a major, poverty-related disease affecting more than 200 million people in developing countries, 85% of them in sub-Saharan Africa. It has a complex epidemiology with a large variation in infection intensity, immune responses to infection, and schistosome-related pathology. Up till now, studies have concentrated on the contribution of host genetics to explain this variation in disease outcome, while the role of parasite genetics remains unexplored. We conducted a large epidemiological study in northern Senegal. We genotyped 1692 *Schistosoma mansoni* larvae collected from 45 human hosts with nine microsatellite loci and linked this with host data such as age, gender, infection intensity, liver and bladder morbidity. Linear regression and redundancy analysis revealed a positive relationship between schistosome infection intensity (measured as eggs per gram feces (epg)), and the frequency of a certain parasite allele. We corrected for age, sex, and co-infection with *S*. haematobium. Two other alleles in this locus were negatively correlated with epg. If we divide infection intensity by worm burden (measured by parasite antigen levels in the blood serum of patients), we have a measure of parasite fecundity. This parameter appeared also positively correlated with the specific allele. The respective microsatellite locus is located in the untranslated region of a protein kinase gene. Inhibiting this gene results in a decrease in schistosome egg production by 30%. Epigenetic control of this gene and its role in host disease phenotype will be discussed.

MAPPING IMMUNE GENE VARIATION ACROSS BOTTLENECKED POPULATIONS OF AN ISLAND BIRD

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Understanding how functional genes evolve is a key issue in evolutionary biology. Genes involved in immune responses are expected to evolve faster than the rest of the genome due to the selective pressure that arises from hosts-pathogen co-evolution. Toll-like receptors (TLR) and beta defensins (BD) have been identified as important effectors of the innate immune response. In vertebrates TLRs function as non-clonal receptors of the immune system that bind and recognize pathogen associated molecular patterns (PAMPs), while BDs directly disrupt the membrane of invading microbes. We investigated the variation at four avian BD loci (AvBD) and five TLR loci in 13 populations of Anthus berthelotii, an endemic passerine that inhabits all islands in the Atlantic archipelagos of Madeira, Selvagens and the Canary Islands. The pipit appears to have gone through a severe population bottleneck when first colonising these islands ca. 75,000 years ago. Now these populations are isolated from each other and have been shown to vary significantly and consistently in terms of the pathogens they harbour. Previous studies have also shown that despite the severe bottleneck variation between populations exists at the major histocompatibility complex (MHC) due to rapid regeneration caused by gene conversion across this gene family. In the present study we found no variation at the AvBD loci, but considerable variation at the TLR loci, both within and across populations. Our evidence suggests that each of the 5 TLR loci are evolving independently, that new haplotypes have been generated since the colonisation but there is no evidence of gene conversion. Analyses of molecular variance show that two TLR loci have significant levels of differentiation between archipelagos. We discuss our findings in the light of the spatially variable pathogen pressures observed across the populations of Berthelot's pipit and in comparison to the variation, and mechanisms of evolution, evidenced at the MHC.

9. Genetics and Genomics of Host-Parasite Coevolution

D21SY09PS1399

NOVEL PAPILLOMAVIRUSES IN FREE-RANGING IBERIAN BATS CHALLENGE THE DOGMAS: INTERSPECIES TRANSMISSION, NO VIRUS-HOST COEVOLUTION AND EVIDENCES FOR RECOMBINATION

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Papillomaviridae are a wide and divergent family of small, non-encapsulated dsDNA viruses that infect most vertebrates. Animal PV diversity is poorly sampled, and thence most of our hypotheses about PV evolution are biased because of the clinical focus on PV research. We communicate here the sequencing and cloning of five new PVs isolated from different bat species: *Eidolon helvum PV type 1* (EhelPV1), Rhinolophus ferrumequinum PV type 1 (RferPV1), Eptesicus serotinus PV type 1 (EserPV1), Eptesicus serotinus PV type 2 (EserPV2) and Eptesicus serotinus PV type 3 (EserPV3). These novel PVs were isolated from three different bat families: Pteropodidae, Rhinolophidae y Vespertilionidae, respectively. Phylogenetic relationships within Papillomaviridae were inferred by means of maximum likelihood, Bayesian, and supernetwork analyses. Further, we have studied the prevalence of EserPV1, 2 and 3 in a number of *E*, serotinus and *E*, isabellinus bat colonies in the Iberian peninsula. There are currently nine bat PVs, and they are not monophyletic: five of them belong to the Lamda+Mu PVs crowngroup, one to the Alpha+Omicron PVs crowngroup, and three of them are close to root and their precise phylogenetic positions cannot be inferred with certainty. The increased sampling of the Lambda+Mu PV crowngroup is welcome, as it hosts viruses infecting diverse hosts (carnivores, chiroptera, primates, rodents and lagomorpha), with diverse tropisms (cutaneous and mucosal) and with divergent clinical manifestations of the infections (asymptomatic, benign proliferations and malignant tumours). Our results question once again the old assumption of virus-host coevolution for the PVs and reinforce the hypothesis of a biphasic evolution in the story of the PVs: an initial adaptive radiation linked to the definition of new niches during the evolution of mammalian skin that generated the ancestral PV crowngroups, followed by a limited co-especiation between viruses and hosts.

TOLL-LIKE RECEPTOR 9 AND HOST DEFENSE TO THE INFECTION CAUSED BY THE HUMAN PARASITE *TRYPANOSOMA CRUZI*

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Toll-like receptor 9 (TLR9) was initially identified as the mammalian component of innate immune system responsible for the recognition of unmethylated CpG motifs of bacterial DNA. We and others have demonstrated the role of TLR9 in infections caused by the human parasite Trypanosoma cruzi. We have shown that this receptor is essential for parasite recognition and host resistance to experimental infections. We have identified CpG motifs in the genome of T. cruzi CL Brener, a hybrid strain. These sequences are not randomly distributed in the genome but instead are enriched clusters containing T. cruzi specific genes. These regions are highly polymorphic in the two haplotypes of the CL Brener strain, which led us to speculate that the abundance of these CpG motifs may vary in the genome of the distinct T.cruzi strains, thus contributing to a differential TLR9 activation. In this work, we studied the role of TLR9 in infections caused by distinct T. cruzi lineages. Experimental infections of wild type and TLR9-/- mice with Colombiana, Y and CL Brener strains revealed a lower importance of TLR9 in controlling parasitemia and mortality in infections with Colombiana, when compared with infections with CL Brener and Y strains. Immunostimulatory assays using live parasites revealed a lower ratio of the TNF- α production by dendritic cells from C57BL/6 and TLR9-/-mice for Colombiana compared with those values for the other strains. These results correlated with the quantification of the regions enriched in CpG motifs in the genome of these strains. Our results suggest that Colombiana infection promotes a lower activation of the TLR9 receptor, leading to insufficient production of proinflammatory cytokines necessary for parasite clearance. We speculate that the content of CpG motifs in the genome of different T. cruzi strains may be an important factor affecting virulence, pathogenesis and favoring parasite adaptation to the vertebrate host.

MULTIPLE-STRAIN INFECTIONS OF BORRELIA AFZELII – A ROLE FOR WITHIN-HOST INTERACTIONS IN THE MAINTENANCE OF ANTIGENIC DIVERSITY?

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Competition is an important evolutionary force and is expected to be most severe between closely related species. In infectious diseases many infections consist of genetically distinct strain, suggesting that and competition between strains might be important. Signs of such competition have been observed in for example malaria infections. Here we investigated the occurrence of competition between closely related strains of the tick-borne disease Borrelia afzelii, one of the species causing Lyme disease in Europe, in one of their most important reservoir hosts, the bank vole (Myodes glareolus). We separated strains based on the highly polymorphic ospC gene using strain specific PCR primers, and found that infected voles on average harbored 2.5 different ospC types. The strains showed a highly aggregated distribution and there were surprisingly little indication of competition. The results rather indicates that strains have a positive effect on each other. Moreover, genetically more different strains were more often found together. In conclusion we suggest that positive interactions might help maintain strain diversity in B. afzelii and that competition between strains play a relatively minor role in this system.

Symposium

10. Genomics and Experimental Evolution

21 August



Program

Wednesday 21 August

Session(s): 5, 6, 7, 8

Organisers: Tadeusz J. Kawecki and Michael G. Ritchie

Invited speakers: Rees Kassen and Thomas Flatt

Description:

This symposium will focus on the application of whole genome resequencing and other high-throughput omics techniques (RNAseq, metabolomics) to experimental evolution, in a broad range of experimental systems. It will facilitate the exchange of information about the questions addressed, techniques adopted and result obtained by different research groups, but also encourage the spread of best practice concerning design and statistical approaches, which are being actively developed in this emerging field.

D21SY10IT10:30R5

GENOMICS OF ADAPTATION IN EVOLVING MICROBIAL POPULATIONS

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The search for the genetic changes responsible for adaptive evolution has been, at least since Mendel, the 'holy grail' of adaptation research. With the introduction of cost-effective next generation sequencing (NGS) technology over the past few years the grail is finally within reach. Combining NGS with experimental evolution of microbial populations is particularly promising in this regard as it provides a glimpse into the natural history of evolving genomes under at least one fairly well defined set of parameters: large population sizes, asexual reproduction, and (usually) haploid genomes. I will review the results of studies that have come from combining NGS with experimental evolution for what they tell us about the genomics of adaptation at the genomic level. In many respects the results are reassuring: the bulk of adaptive changes occur in open reading frames and are non-synonymous, for example. But NGS has also provided some surprises: synonymous mutations that are clearly adaptive, for example, and mutations in genes that, at first glance at least, are hard to interpret in an adaptive light. Making sense of the full spectrum of results will require some careful rethinking in terms of experimental design and genomic sampling.

D21SY10IT14:00R5

GENOMIC PATTERNS OF EXPERIMENTAL EVOLUTION OF DROSOPHILA LIFESPAN

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The genomic basis of molecular polymorphisms that underlie adaptive phenotypic differentiation remains poorly understood. A potentially very powerful approach towards resolving this fundamental problem is to combine experimental evolution with next generation sequencing (NGS). In the last two to three years, a handful of studies have begun to combine experimental evolution in Drosophila with NGS in order to uncover the genomic basis of phenotypic differentiation due to laboratory selection. In the first part of my talk, I will briefly review these recent efforts and highlight interesting aspects of their methods and results. In the second part of my talk, I will focus on the genomic basis of artificial selection for increased lifespan and late-life fertility in Drosophila. The first artificial selection experiments on Drosophila lifespan were carried out about 30 years ago by Michael Rose and Brian Charlesworth and, independently, by Leo Luckinbill and Robert Arking and collaborators. In 2011, Remolina et al. have performed the first analysis of genome-wide changes in response to 50 generations of artificial selection for increased lifespan in Drosophila. In their experiment, the authors identified many interesting longevity candidate genes that are associated with oogenesis, immunity, and proteolysis. I will compare their novel findings to our preliminary analysis of the genomes of the Luckinbill/Arking lines, which by now have undergone approximately 150 generations of laboratory selection for postponed senescence. I will address the key question of whether our study turns up the same candidates and pathways as found by Remolina and colleagues, or whether most candidates do not overlap, maybe because there exist many different genetic ways to evolve the same life history phenotype.

D21SY10RT11:18R5

TRAMPLE THE WEAK - PHENOTYPIC AND GENOMIC EVOLUTION OF PSEUDOMONAS AERUGINOSA IN IMMUNOCOMPROMISED HOSTS

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Novel infectious diseases have been emerging at an alarming rate over the last few decades, with sometimes devastating consequences for public health. Prime examples include the rise of HIV or the increasing incidences of tuberculosis and MRSA. To understand the origin of these novel strains, detailed knowledge is required on the adaptive evolution underlying successful establishment and spread of emergent strains in new host populations. Both theoretical studies and occasional epidemiological reports identified host immune state as a key factor in disease emergence. However, the importance of this factor has not been studied in a rigorous experimental framework that explicitly allows for long-term evolution. I will present the results of an evolution experiment in which Pseudomonas aeruginosa PA14 adapted to populations of immunocompetent C. elegans and immunocompromised (mutant) host strains. Phenotypic analyses revealed rapid, to a large extent parallel evolution across all treatments. Strong increases in bacterial fitness were especially observed in immunocompromised hosts, suggesting immunocompromised hosts facilitated faster bacterial adaptation. Genomic analyses confirm the observed phenotypes and the large amount of parallel evolution. However, they further revealed distinct evolutionary paths chosen by bacteria adapting to immunocompromised hosts compared to those adapting to immunocompetent hosts. These results provide important insights for the understanding of bacterial adaptation to host populations, and may assist the development of theoretical models on pathogen evolution and adaptation.

D21SY10RT11:42R5

EXPERIMENTAL EVOLUTION OF ESCHERICHIA COLI UNDER INEFFICIENT SELECTION REVEALS THE EVOLUTIONARY TRAJECTORIES OF SYMBIOTIC BACTERIA

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Many bacteria that establish mutualistic associations with insects are transmitted from mother to offspring under a vertical regime. This transmission mode imposes strong bottlenecks on the effective population sizes of bacteria, resulting in a high genetic drift effect and, consequently, increased genetic variability among populations and decreased polymorphism within populations. Inactivation and degradation of genes that are unnecessary in an intracellular environment is a common symptom of symbiotic lifestyle. Two main questions remain unanswered with regards to the evolution of these bacteria: (a) how do essential genes for symbiosis counteract the effect of mutations fixed under inefficient selection in the genome? And (b) what mechanisms increase the mutational robustness of symbiotic genes? Here we have conducted an evolution experiment using the bacterium Escherichia coli to simulate the mutation dynamics under inefficient natural selection occurring in symbiotic bacteria. Evolution of E. coli for more than 5000 generations reveals the network of interactions between mutations, the set of useful, although not essential, genes for symbiosis and the role of GroEL in mutational robustness. Our results unveil a more complex scenario for the evolution of symbiosis than anticipated by previous studies.

D21SY10RT14:48R5

GENOMIC CHANGES UNDER RAPID EVOLUTION: SELECTION FOR PARASITOID RESISTANCE

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Most studies of selection in experimental populations so far have focused on long term selection pressures of relatively small effect. In this study we use the parasitoid Asobara tabida as a strong, fast and ecologically relevant selection pressure on four replicate selection lines derived from an outbred, natural population of *Drosophila melanogaster*. Parasitoids are insects whose larvae feed on a host in order to complete development and in this process kill the host. In the Drosophila genus the ability to successfully induce an immune response to encapsulate and melanize a parasitoid egg, is present in the melanogaster subgroup, where *D. melanogaster* shows a high level of variation in immune defense. By combining artificial selection and whole genome HT-sequencing we identified several genomic regions carrying a signature of selection. In these selected regions we selected candidate genes associated with one or more significant SNPs, which potentially confer increased resistance against parasitoid attack. We propose that a 600kb region on chromosome 2R is a region of major effect on parasitoid defense, as it is highly enriched in highly significant SNPs. The power of our approach is that we use replicated artificial selection experiments and a natural selective agent on an outbred *D. melanogaster* strain. This enables us to show very narrow genomic ranges that have been affected by selection, even under such a fast and strong selective regime. In addition to providing a much more detailed insight in how rapid evolution impacts the genome, it has also given us specific hypothesis on the genes and genomic regions that confer higher defenses against parasitoids.

D21SY10RT15:12R5

UNDERSTANDING THE RELATIONSHIP BETWEEN NATURAL AND SEXUAL SELECTION USING THE EVOLVE AND RESEQUENCE APPROACH IN A NON-MODEL INSECT

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Sexual selection is a source of strong directional selection in nature that has received long-standing attention due to its role in the evolution of elaborate sexual ornaments and armaments. Although its contribution to the evolution of sexual traits is uncontroversial, the way in which sexual selection works alongside natural selection, specifically, whether the process favors similar or different allelic variants to natural selection, remains poorly understood. To address this question we assessed the genome-wide response from standing variation to natural and sexual selection using experimental evolution in a fly. RAD-seq sequencing was applied to replicate experimental populations of Drosophila serrata that were allowed to adapt to a novel environment over sixteen generations. Each line was randomly assigned to one of four experimental treatments that reflected a full factorial manipulation of the opportunity for natural and sexual selection. Our analysis of the genomic response suggests that the interaction between natural and sexual selection can lead to contrasting allele frequency trajectories compared to when either process operates alone. In the course of outlining our study I will address some of the statistical challenges posed by the analysis of "evolve and resequence" studies that are short-term and often involve highly polygenic responses. This will include our application of multivariate tools and generalised linear mixed effects models to experimental evolution data.

D21SY10RT15:45R5

THE IMPACT OF CHROMOSOMAL INVERSIONS ON GENETIC VARIATION IN NATURAL AND LABORATORY POPULATIONS OF D. MELANOGASTER

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Paracentric inversions are very common structural variants in many species of the genus Drosophila. Despite a large body of literature, still very little is known about the genetic mechanisms underlying inversion polymorphisms. Recent advances in sequencing technology now allow studying the distribution of genetic variation on a genome-wide level, which has reignited interest in the genomic basis of inversion evolution. To extend previous efforts we have combined karyotyping of polytene chromosomes with whole-genome sequencing in order to identify polymorphic inversions and examine associated haplotype structure in populations derived from a laboratory natural selection experiment in D. melanogaster. In addition, we combined information from 110 D. melanogaster genomes of known karyotype from Africa, Europe and North America to investigate patterns and distribution of fixed differences linked to different inversions. We used this novel dataset to estimate inversion frequencies from pooled next generation sequencing data ("Pool-Seq") in our selection experiment and in populations collected along the North American and Australian latitudinal cline. Among six polymorphic inversions segregating in the experimental populations, two rare cosmopolitan inversions, In(3R)C and In(3R)Mo, showed a frequency increase consistent with non-neutral evolution. Genetic variation in and around *In(3R)Mo* was strongly reduced, consistent with previous findings from North America, and we found evidence for gene flux between this inversion and the non-inverted standard arrangement. Moreover, we identified a previously unknown latitudinal cline for *In(3R)Mo* in our Pool-Seq data from the North America east coast. Our novel data highlight the impact of inversions on patterns genetic variation and underline the importance of considering structural variants when attempting to detect patterns of adaptation.

D21SY10RT16:09R5

GENETIC BASIS OF AGEING EVOLUTION UNDER DIFFERENTIAL EXTRINSIC MORTALITY IN A NEMATODE

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Ageing is inevitable in most living organisms but how ageing evolves is unclear. Because of extrinsic mortality due to a variety of biotic and abiotic factors, the strength of selection declines with age. This "selection shadow" may result in the accumulation of detrimental mutations with late-life expression or fixation of antagonistically pleiotropic mutations that increase fitness in early-life at the expense of fitness in late-life. Classic theory has thus predicted that if mortality is high, the evolutionary contribution of old individuals will be low and that this will lead to evolution of accelerated ageing and decreased longevity. However, if mortality is non-random, condition-dependent selection is likely to yield individuals with strong resistance to typical causes of mortality. Emerging theory thus suggests that condition-dependence may alter, and even reverse, the classic pattern. We have previously employed an experimental evolution design, using a nematode worm Caenorhabditis remanei, that allowed us to disentangle the effects of mortality rate (high vs low) and mortality source (random vs heat-shock) on the evolution of longevity. We observed the evolution of reduced longevity under high random mortality, confirming the classic prediction. In contrast, high condition-dependent mortality led to the evolution of increased longevity and lower late-life mortality rates, supporting a key role for mortality source in the evolution of ageing. In the present project, we examine which genes underlie the evolution of lifespan by quantifying divergences in gene expression across our selection lines. Using RNA sequencing, we quantified the differentially expressed genes in young and old nematodes from the four selection regimes. The results will be discussed.

D21SY10RT16:33R5

EFFECTS OF EXPERIMENTAL MATING SYSTEM VARIATION ON TRANSCRIPTOME EVOLUTION AND MATING RESPONSE IN FEMALE DROSOPHILA PSEUDOOBSCURA

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Interactions between the sexes are believed to be a potent source of selection on sex-specific evolution, but the way in which sexual interactions influence females at the molecular level is poorly understood. We examined the effects of 100 generations of elevated polyandry and enforced monandry on gene expression evolution in female *Drosophila pseudoobscura*. We assessed differences in gene expression between females from the two selection regimes and how the expression response to mating depended on female sexual selection history, indirect genetic effects (IGEs) due to the genotype of the male and female-by-male genotype interactions. Our data show large-scale gene expression divergence between the experimentally evolved females, with selection mainly targeting female-biased genes expressed in the ovaries. Most of the expression response to mating between the experimental females. The indirect genetic effects of the male selection history on the female gene expression response were surprisingly small. Our results provide critical experimental evidence for a role of female-specific selection arising from polyandry in promoting rapid evolution of the female transcriptome.

D21SY10RT16:57R5

EVOLUTION UNDER MONOGAMY FEMINIZES GENE EXPRESSION

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Many genes have evolved sexually dimorphic expression as a consequence of divergent selection in males and females. However, the degree to which evolution can shape gene expression independently in each sex is controversial. It is possible to directly test whether genetic constraints have prevented sex-specific optima from being obtained by enforcing a monogamous mating system, which eliminates both female choice and male-male competition. This relaxed selection on males should result in evolution towards female-specific optima for any genes previously under antagonistic selection in males. Here we demonstrate broad constraints on the evolution of sexually dimorphic gene expression by maintaining populations of Drosophila melanogaster for 65 generations under strict monogamy. Compared to polygamous controls, monogamous populations evolved lower expression of male-biased transcripts and higher expression of female-biased transcripts. This pattern of feminization was present in both sexes, evident throughout the genome and across tissue types, and strongest for genes located on the X chromosome, a known hotspot for sexually antagonistic selection is ubiquitous in the genome and likely to play a major role in maintaining genetic variation, driving the evolution of sex chromosomes, and reducing population-level fitness.

D21SY10RT17:45R5

GENOMIC SCOPE OF ADAPTIVE MUTATIONS TO DIFFERENT ENVIRONMENTS

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A panel of adaptive mutations was obtained by exposing Saccharomyces cerevisiae strains to harsh environmental conditions. The genomic scope of mutations allowing adaptation to these conditions was then determined by sequencing 35 different adapted lines from each of two environments. This panel was then used to measures trade-offs in different environments, as well as properties such as dominance across environments. The implications for Haldane's sieve and the role of ploidy in adaptation will be discussed.

D21SY10RT18:09R5

SHIFTING FITNESS LANDSCAPES IN RESPONSE TO ALTERED ENVIRONMENTS

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One of the most controversial questions in evolutionary biology is the role of adaptation in molecular evolution. Following decades of debate between selectionists and neutralists, new high-throughput methods are beginning to illuminate the full distribution of fitness effects of new mutations. Here, we shed light on the adaptive potential in *Saccharomyces cerevisiae* by presenting systematic high-throughput fitness measurements for 578 possible point mutations in a region of Hsp90 in each of six environmental conditions. Under elevated salinity, we observe numerous beneficial mutations with growth advantages up to 7% relative to the wild type. We thus demonstrate that an essential protein can harbor adaptive potential upon an environmental challenge, and report a remarkable fit of the data to Fisher's geometric model - which well predicts the observed costs of adaptation associated with environmentally specific beneficial mutations. Furthermore, we utilize a Bayesian MCMC modeling framework to evaluate the statistical significance of the results - showing a remarkable accuracy of the experimental approach, e.g., allowing for direct inference of the non-neutrality of certain synonymous substitutions.

D21SY10RT18:33R5

THE GENETIC BASIS OF ADAPTATION TO OCEAN ACIDIFICATION IN EMILIANIA HUXLEYI , A KEY PHYTOPLANKTON SPECIES

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Ocean acidification, the drop in seawater pH due to the uptake of anthropogenic CO2, is a major threat to marine calcifying organisms. In a long-term evolution experiment, we found that the biogeochemically important unicellular algae *Emiliania huxleyi* partly restored calcification and growth rates at elevated CO2 owing to adaptive evolution, compared to control lines kept at ambient CO2 levels. In order to unravel the genetic basis of adaptive change we re-sequenced replicated control and adapted populations to a high coverage. We identified SNPs and other polymorphisms that diverged with respect to CO2 selection treatments via mapping to the existing genome of *E. huxleyi*. We were particularly interested whether or not the same genes or metabolic pathways were affected, and how consistent these changes were across replicates. These data provide first insights into the dynamics and the parallelism of de novo mutations in an ecologically important phytoplankton species.

D21SY10RT18:57R5

GENOMIC ADAPTATION TO DIFFERENT HOSTS - WHAT MAKES BETTER-ADAPTED VIRUSES?

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The interplay between ecological and genomic adaptation is at the heart of evolutionary processes. If we consider that species evolve as groups of genomes adapting to particular ecological niches, shifts to a new ecological niche should be connected with the adaptation and differentiation of a genome population to this particular niche. To study how ecological adaptation affects genomes simultaneously at both ecological and whole genome levels, we focussed on large DNA viruses because their ecological niches are mostly defined by their hosts. Among viruses, baculoviruses are uniquely transmitted between insect hosts as groups of genomes, which sets them further apart for studying the effect of niches on populations. We investigated this question of ecological genomic adaptation by an experimental evolution approach on a baculovirus. As their infectious particles contain >100 virions each enclosing a genome of 134 kb, baculoviruses foster the maintenance of high genetic diversity, as we discovered from ultra-deep illumina sequencing of the AcMNPV wild-type isolate. We found different level of polymorphism: few SNPs at high frequency located in essential genes involved in replication, transcription and transmission, and low frequency SNPs in every position, suggesting any potentially beneficial mutation is already present within the population. We used this diverse virus population to perform experimental evolution in two lepidopteran host species Trichoplusia ni and *Spodoptera exiqua*. We performed 10 infection cycles in 10 replicate lines. We analysed the evolution of virulence phenotypes in parallel to resequencing the evolved progeny lines.

POSTERS

D21SY10PS0098

EXPERIMENTAL THERMAL EVOLUTION IN D. MELANOGASTER REVEALS TEMPERATURE DEPENDENT POPULATION GENOMIC SIGNAL OF ADAPTATION

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How adaptation effects segregating variation at the population genomic level in sexually reproducing diploids remains a poorly understood yet fundamental biological question. However, recent advances in sequencing technology in combination with experimental evolution have promised to reveal the temporal patterns of genomic adaptation down to individual SNP resolution. Here I discuss results from 15 generations of experimental evolution in replicated populations of *D. melanogaster* maintained in two separate thermal environments, which mimic either heat and cold stress. When taking the top candidate SNPs from each base-evolved population comparison, we find an enrichment of hot candidate SNPs in genes associated with heat tolerance, and likewise for cold candidate SNPs in cold tolerance genes, but not vice versa. Furthermore, we find that the rising allele (i.e. that most likely to be under selection) tends to start at either low or intermediate frequencies in the hot and cold treatments, respectively. Hence, it appears that thermal selection is involved in driving changes between the two treatments and is deferentially dependent on the starting allele frequency. The possible causes behind these intriguing patterns are discussed with respect to our emerging understanding of thermal adaption in *D. melanogaster*.

D21SY10PS0157

EXPERIMENTAL EVOLUTION AND TRANSCRIPTIONAL PLASTICITY IN AN INSECT-FUNGUS INTERACTION

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Organisms rarely occur solitary; viewed in this light, interaction is forced upon them. The fruit fly Drosophila melanogaster, frequently used as a model organism in evolutionary studies, makes use of rotting fruits as breeding sites. Concurrently, filamentous fungi utilise rotting organic material to obtain their nutritional substances. One trait of many fungi is the production of toxic secondary metabolites. It is hypothesised that these toxins hold a function as a chemical defence against other microbes, e.g. antibiotics like *Penicillin*, but furthermore features also a defence against larger organisms like insects. Drosophila larvae are constrained in leaving a food patch, due to the risk of desiccation, and have evolved other traits to cope with noxious fungi and their toxins on infested substrates. In an experimental-evolution approach, we mimic the infestation of *Drosophila* habitats by the filamentous fungus Aspergillus nidulans. A wild-type and a toxin-production-impaired mutant strain, as well as the mycotoxin Sterigmatocystin are used as selection pressures. After multiple confrontations, over 25 generations, we will analyse changes in the transcriptional response of the fly larvae using RNAseq. Additionally, changes in the immune respons and in phenotypic traits will be recorded. Currently, we are analysing the transcriptional plasticity of the fly larvae by disentangling the response towards the fungus and the mycotoxin. For this purpose we have confronted fly larvae with the 3 treatments (WT, mutant, toxin) that are used in the selection experiment. Over a time series, samples were taken and sequenced through NGS. We will compare this, yet unknown, larval response to the transcriptional response after experimental evolution of increased tolerance and/or resistance against the noxious fungus. Here we will present our current results on the dynamics of the interaction between Drosophila larvae and the toxin-producing fungus A. nidulans.

D21SY10PS0182

HIGH RESOLUTION LINKAGE ANALYSIS DURING EXPERIMENTAL EVOLUTION EXPERIMENTS

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Experimental evolution in combination with whole genome resequencing is a promising approach to investigate evolutionary responses to direct selective treatments or novel environments. In particular *D. melanogaster* has been used as a model for a sexually reproducing diploid organism. Using Pool-Seq to identify putative targets of selection, all studies identified a vast number of putatively selected loci. Hence, it appears likely that the distinction of true and false positives is a problem common to these studies. Most important, false positives do not necessarily appear only in close proximity to the targets of selection, but appear to be distributed throughout the entire genome. Reasoning that more detailed information about dynamics of the underlying haplotype structure will be highly instrumental to distinguish true and false positives, we analyzed the patterns of linkage disequilibrium in an experimental evolution study with *D. melanogaster* adapting to a novel temperature regime. We will show how the information about whole chromosome linkage pattern changes during experimental evolution and to what extent this linkage pattern provides the basis of an improved strategy to distinguish between true and false positives in Pool-Seq data from experimental evolution studies.

D21SY10PS0183

RAPID GENOMIC CHANGES IN PATHOGENIC BACILLUS THURINGIENSIS DURING ADAPTATION TO ITS NEMATODE HOST

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Bacillus thuringiensis is a pathogenic bacterium of invertebrates with a wide host spectrum, including the nematode Caenorhabditis elegans. We studied its ability to adapt to the nematode host with the help of an evolution experiment, during which a mixture of B. thuringiensis strains was either coevolved with C. elegans (coevolution treatment), adapted to a non-changing host population (one-sided adaptation), or evolved in the absence of the host (control evolution). Experimental evolution produced distinct phenotypic changes in virulence and also other life-history traits. Here, I present results on our analysis of the genetic basis of evolutionary changes based on whole-genome sequencing of replicate populations. Firstly, we were able to identify a central role for clonal selection during experimental evolution, especially under coevolution and control conditions. Each of these conditions was dominated by two different chromosomal genotypes, whereas an unexpected variability among replicate populations was observed in the one-sided adaptation treatment. Secondly, we were able to demonstrate that adaptation to the host is additionally influenced by the spread of genomic SNP and indel alleles as well as changes in plasmid composition and presence of phage infection. Taken together, our results highlight that the trajectory of evolution depends on multiple interaction levels: (i) host – pathogen; (ii) competition between pathogen strains and (iii) selfish mobile elements comprised of plasmids and phages.

D21SY10PS0199

STUDYING THE EFFECTS OF PREDATION AND SEX ON THE EVOLUTION OF PREY DEFENSE TRAITS IN CHLAMYDOMONAS REINHARDTII

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Understanding the evolutionary advantage of sexual reproduction, as well as its maintenance and widespread occurrence, remain key areas of focus within evolutionary biology. Given the associated costs of sex, including the break-down of potentially beneficial genetic associations via recombination and the time-consuming process of finding a suitable mate, the fundamental question "why sex?" persists. Despite this, complete asexuality in the eukaryotic kingdom is rare and a substantial number of species rely on sex as the sole mode of reproduction. Previous studies have shown sex to confer an advantage in populations exposed to novel environments or intense selective stressors, as recombination can increase genetic variation, thereby facilitating increased rates of adaptation. Asexual populations may reproduce faster, but must then rely on standing genetic variation, which may be very low, and/or novel mutations to supply variation necessary for adaptation, which may happen slowly or not at all. Theoretical models based on these ideas require empirical validation; an effective research framework to study such evolutionary processes in real time would be to combine experimental evolution with ecogenomic analyses as a way to test for adaptive evolution and link phenotype with genotype for ecologically-relevant traits. The purpose of this project is to carry out such a study by using the predator-prey model system of rotifers, *Brachionus calyciflorus*, and unicellular green alga, Chlamydomonas reinhardtii, to test for adaptive evolutionary responses to rotifer predation in both sexual and asexual lines of alga. Through the use of next-generation sequencing technology and bioinformatics, we aim to better understand the underlying genetic basis of adaptive traits, with respect to defense, in *C. reinhardtii*. In turn, these results may help to elucidate the evolutionary consequences of sexual versus asexual reproduction, and we can better answer, "Why sex?"

D21SY10PS0450

EXPERIMENTAL EVOLUTION OF FEMALE TRAITS UNDER DIFFERENT LEVELS OF INTER SEXUAL CONFLICT

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A number of studies have documented the evolution of female resistance to mate harm in response to the alteration of intersexual conflict in the populations. However, the life-history consequence of such evolution is still a subject of debate. In the present study, we subjected replicate populations of Drosophila melanogaster to different levels of sexual conflict (generated by altering the operational sex ratio) for over 45 generations. Our results suggest that females from populations experiencing higher level of intersexual conflict evolved increased resistance to mate harm, in terms of both longevity and progeny production. These females were also found to have increased locomotor activity, a possible correlated response to the selection on males in that regime. Females from the populations with low conflict were significantly heavier at eclosion and were more susceptible to mate harm in terms of progeny production under continuous exposure to the males. However, these females produced more progeny upon single mating and had significantly higher longevity in absence of any male exposure – a potential evidence of trade-offs between resistance-related traits and other life-history traits, such as fecundity and longevity. We also report tentative evidence, suggesting an increased male cost of interacting with more resistant females.

D21SY10PS0511

A NOVEL FUNCTIONAL POLYMORPHISM IN THE FATTY ACID DESATURASE 2 GENE (FADS2): POSSIBLE ROLE IN BASAL METABOLIC RATE

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As membrane components, polyunsaturated fatty acids (PUFAs) play an important role in cellular processes and have been shown to be associated with basal metabolic rate (BMR). While the link between BMR and membrane lipid composition is clear on an interspecific level, the underlying mechanism linking them on an intraspecific level is not well understood. Probably one reason for this is that variation of a physiological trait like BMR is much narrower within a species than between organisms from two different species, and in turn the mechanisms explaining any observed variation could be subtler. In this circumstance, a much stronger methodology for studying BMR should be found in artificial selection experiments which manipulate the frequencies of genes directly related to the trait under study. Here we describe a new polymorphism in the fatty acid desaturase (Fads2) gene for Δ -6 desaturase (D6D), a key enzyme for PUFA synthesis, in 120 males from two genetic lines of outbred Swiss Webster laboratory mice (Mus musculus) selectively bred for 32 generations (F32) towards low and high BMR. For controlling the force of genetic drift, we also used 78 mice of generation 22 (F22). For the same reason, we analyzed 36 mice from three unselected lines of mice maintained in a separate, concurrent selection experiment in generation 16 (F16). The aim of this study was to verify a possible association between Fads2 genotypes and BMR. We found significant correlation between Fads2 variants and BMR within selected, as well as unselected lines of mice. We also used a FST outlier test to asses the effect of selection on the variation between L-BMR and H-BMR lines of mice at the Fads2 gene, using the Fads2 gene, together with data from the 11 microsatellite loci. Taken together, these results suggest that Fads2 is expected to become a major focus of membrane, metabolic rate and human metabolic syndrome research.

D21SY10PS0584

FROM ARTIFICIAL SELECTION TO TRANSCRIPTOMICS: THE COGNITIVE BENEFITS OF, AND THE GENE RESPONSIBLE FOR, A LARGE BRAIN IN FISH

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The evolutionary causes and consequences of the enormous phenotypic variation in brain size among vertebrates remain enigmatic. Positive selection for increased cognitive abilities is a likely driver of brain size evolution, but the genetic bases of vertebrate brain size variation is largely unexplored. An experiment linking brain size, cognition and gene expression differences is therefore lacking. We used artificial selection for large and small brain size relative to body size in the guppy (Poecilia reticulata) to experimentally investigate the link between brain size, cognitive ability, and its genetic background. We found that brain size evolved rapidly in response to divergent selection, and that large-brained females outperformed small-brained females in a numerical learning assay designed to test cognitive ability. Whole transcriptome sequencing (RNA-Seq) of the brain of large- and small-brained fish revealed a dramatic expression difference in a single gene, Angiopoietin-1, which is an endothelial growth factor highly conserved among vertebrates and known to promote both neural proliferation and neural differentiation. We therefore show that a large brain size indeed confers cognitive benefits and nominate Angiopoeitin-1 as the candidate gene mediating the evolution of brain size and cognitive ability in vertebrates.

D21SY10PS0681

DETECTION OF TRACES OF SELECTION WITH NUMEROUS SNP IN SMALL EXPERIMENTAL POPULATIONS UNDERGOING DIRECTIONAL SELECTION

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Three lines of chickens (L1, L2, L3) have been selected for 12 generations for three different immune response traits. A fourth line (L4) was a contemporary random bred Control (Minozzi et al. 2008). Each generation, 200 chicks per line were hatched in a single batch. Selection for each trait was done by mass selection based on individual phenotype. Individuals from the three selected lines and the control line at generation G9, as well as individuals from the founding population (G0) were sampled (20 individuals/line) and genotyped with a 60K SNP chip. We present the use of this dataset to detect traces of selection by comparing the SNP allele frequencies between generations G0 and G9 for each line. The method was able to pinpoint a dwarfing gene known to have undergone strong selection hence, serving as a validation. In addition it highlights numerous SNPs that seem to behave non-neutrally, providing candidate regions for future search for selected genes. While classical approaches generally focus on traces of 'historical' (long term) selection, this work demonstrates that it is possible to detect short-term selection in experimental population using SNPs. Minozzi, G, et al., BMC GENETICS, 9:5, 2008.

D21SY10PS0718

EXPERIMENTAL EVOLUTION AND TRANSCRIPTOMICS OF TRIBOLIUM CASTANEUM INFECTIONS WITH BACILLUS THURINGIENSIS

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Experimental evolution of hosts and parasites can help to elucidate the genomic basis of fast evolutionary processes. We are therefore using experimental coevolution in the red flour beetle *Tribolium castaneum* and its microparasite *Bacillus thuringiensis* to address evolving genetic specificity, while experimental selection on the hosts' immune system addresses the evolution of immunological specificity. We use two different infection routes, the oral route of infection, and infection through septic prick injury, which brings the bacterium into direct contact with the host's immune system. Through RNA sequencing we characterised the genomic responses of the host to these two infection routes. We found astonishingly distinct transcriptomic responses for oral versus septic infection. To test for genetic differences between host populations we compared the transcriptomic responses to infection. Not only is the latter population more resistant upon oral infection, but we also found that a much higher number of genes were differentially expressed after oral infection. Studying the transcriptomes of the evolved hosts will thus shed further light on the genomic basis of host-parasite coevolution.

D21SY10PS0752

EDAPHIC ADAPTATION IN THE 'OMICS' ERA: A GENOMIC VIEW OF THE CALCICOLE-CALCIFUGE PROBLEM

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Since almost 200 years, plant ecologists are debating about the causal factors underlying the striking floristic differences observed between plant communities occupying calcareous versus siliceous substrates. Indeed, it remains unclear which chemical (e.g. aluminium and bicarbonate toxicity, nitrate or phosphate depletion) or physical factors (e.g. water or temperature stress) actually drive edaphic adaptation in natural plant populations, advocating for comparative genomic studies on calcicolecalcifuge vicariants, i.e. sister taxa that grow on either soil type. We herewith propose to fill this knowledge gap, by deciphering the genetic basis of local adaptation in two diploid species of Arabidopsis (A. lyrata and A. arenosa, Brassicaceae) that grow on both calcareous and siliceous bedrocks. Using whole-genome re-sequencing of pooled DNA libraries (totalling 240 individuals per species), along with outlier analyses based on allele frequency divergence, we have identified a hundred loci that are enriched for genes acting at the cell periphery in response to ion homeostasis and trans-membrane transporter activity. Among them, a dozen contain up to 130 outlier SNPs, and constitute excellent candidates for functional validation. Overall, our results suggest that chemical soil composition is the main cause of ecological differentiation in these species. Our work further highlights the potential of *Arabidopsis* as a new model system in experimental studies on edaphic adaptation in plants.

D21SY10PS0773

EVOLUTION OF SPERM COMPETITIVE ABILITY AS A CORRELATED RESPONSE TO SELECTION ON ENVIRONMENTAL STRESS RESISTANCE

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Sperm competitive ability in Drosophila melanogaster is a crucial component of male fitness. Few empirical studies have focused on the effects of environment on evolution of sperm competition. We evolved laboratory population of Drosophila melanogaster to resist cold shock. After 11 generation of selection we found that the selected population eggs had higher hatchability post cold shock. Interestingly, following a cold shock, males from the selected populations were able to induce previously mated females to mate again. They also sired more progeny indicating better ability of sperm competition. There results indicate that sperm competition can evolve rapidly under adaptation to environmental stresses.

D21SY10PS0986

POPULATION AND SEX DIFFERENCES IN DROSOPHILA MELANOGASTER BRAIN GENE EXPRESSION

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Changes in gene regulation are thought to be crucial for the adaptation of organisms to their environment. Transcriptome analyses can be used to identify candidate genes for ecological adaptation, but can be complicated by variation in gene expression between tissues, sexes, or individuals. Here we use high-throughput RNA sequencing of a single Drosophila melanogaster tissue to detect brainspecific differences in gene expression between the sexes and between two populations, one from the ancestral species range in sub-Saharan Africa and one from the recently colonized species range in Europe. Relatively few genes (<100) displayed sexually dimorphic expression in the brain, but there was an enrichment of sex-biased genes, especially male-biased genes, on the X chromosome. Over 340 genes differed in brain expression between flies from the African and European populations, with the between-population divergence being highly correlated between males and females. The differentially expressed genes include those involved in stress response, olfaction, and detoxification. Expression differences were associated with transposable element insertions at two genes implicated in insecticide resistance (Cyp6g1 and CHKov1). Analysis of the brain transcriptome revealed many genes differing in expression between populations that were not detected in previous studies using whole flies. There was little evidence for sex-specific regulatory adaptation in the brain, as most expression differences between populations were observed in both males and females. The enrichment of genes with sexually dimorphic expression on the X chromosome is consistent with dosage compensation mechanisms affecting sex-biased expression in somatic tissues.

D21SY10PS1124

LOSS OF HETEROZYGOSITY DURING ASEXUAL REPRODUCTION IN DAPHNIA

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Explaining the ubiquity of sexual reproduction despite its high costs remains one of the major challenges in evolutionary biology. This problem is known as "paradox of sex" and many theoretical models concerning it have been proposed. Following the recent breakthroughs of sequencing technologies we are now able to experimentally evaluate some of the basic assumptions of these models. We used Restriction site Associated DNA (RAD) sequencing to examine the common assumption that asexual reproduction results in perfect clonal offspring with mutations being the only source of new variation. For this we chose the emerging model system *Daphnia magna*, a cyclic parthenogenetic planktonic crustacean. Comparing pathenogenetic offspring with their mothers at several thousand genetic markers, we detected loss of heterozygosity (LOH) within single asexual generations of *D. magna*. Our results indicate surprisingly high rates of genome homogenization events (homologous recombination, gene conversions or ploidy losses) during asexual reproduction. These findings should be incorporated in future theoretical models which might lead to the resolution of the "paradox of sex".

D21SY10PS1136

TRANSCRIPTOMICS OF ACCESSORY GLAND PROTEINS IN SEX PEPTIDE RECEPTOR-LESS DROSOPHILA MELANOGASTER POPULATIONS

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Accessory gland proteins (Acps) are male reproductive tract proteins transferred to females during copulation, which alter their reproductive physiology and behaviour. One of the Acps in Drosophila melanogaster, sex peptide (SP) not only alters fecundity and sexual receptivity of the females, but also affects their immune system, feeding, and longevity. It is thus sexually antagonistic and costly to the females. We want to examine whether production of SP is costly to the males. SP acts through binding to specific sex peptide receptors (SPR) in the female reproductive tract and the central nervous system. We are studying gene expression levels of SP in males from selection populations of *D. melanoqaster* in which females do not express the sex peptide receptor. The benefit of producing SP being thus removed, we hypothesize that, if production of SP is costly to males, this should lead to a decreased gene expression of SP in such populations. We also intend to test whether the males compensate by increasing production of other Acps/seminal proteins such as Ovulin and DUP99B (Ductus ejaculatorius peptide 99B) since these proteins have some overlapping functions with SP. RT-qPCR will be used to quantify the expression of SP, Ovulin and DUP99B in the males. ELISA of female reproductive tract will be used to quantify the Acps transferred to the females during mating. This will indicate whether or not there is a correlation between gene expression levels and the amount of protein retained in the females an hour after mating.

D21SY10PS1137

MEIOTIC DRIVE IN THE YELLOW FEVER MOSQUITO AEDES AEGYPTI

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Selfish genetic elements enhance their own transmission to the organism's progeny in a disproportionate fashion. Sex chromosome meiotic drive normally involves X chromosomes and results in female biased sex ratios, but in the yellow mosquito *Aedes aegypti* a meiotic drive gene is tightly linked to the male chromosome and distorts the sex ratio towards males. This distortion can only be seen when males bearing the meiotic drive gene (D) are crossed to females from other populations. That is because a dominant insensitive gene (I) has evolved in females of the populations where D is present, preventing the sex ratio distortion while females from other geographical regions are commonly sensitive (s) to D. We crossed females of a line that is highly sensitive to meiotic drive to a line carrying the drive gene. Using restriction-site associated DNA (RAD) tags followed by deep sequencing, we were able to both map the meiotic drive gene and genes elsewhere in the genome which suppress meiotic drive. We then backcrossed the meiotic drive gene into a genetic background without any suppressors and repeated the experiment. This allowed us to map the meiotic drive gene with greater precision.

D21SY10PS1333

THE RATE OF ADAPTATION ESCHERICHIA COLI TO THE MOUSE GUT

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The process of adaptation in bacterial populations is often studied in simple and well-defined laboratory environments, mainly involving abiotic interactions. On the other hand, adaptation to complex environments, within ecological communities, involving biotic interactions, is only rarely studied. A medically important and extremely diverse community is the gut microbiome. Here, we study the process of adaptation of *Escherichia coli* to the mouse gut. We combine methodologies from evolutionary genetics and molecular biology to understand the rate of adaptation and the number and selective strength of newly arising mutations. Previous work from our lab has shown that, in contrary to what is theoretically predicted and normally observed in-vitro, the rate of adaptation does not decrease through time, during two consecutive colonizations of the mouse gut (~900 generations). We now isolated three genetically distinct clones (from the second colonization), which represent three independent evolutionary paths and test whether the evolutionary process proceeds at the same pace (both in terms of rate of adaptation and selective strength of new mutations). To better understand the adaptive process, we use genome re-sequencing of the evolved clones and measure their fitness across a variety of environments that mimic different components of the gut environment. Our results, will allow us to further test the relevance of the following theoretical predictions: 1) as populations adapt to a given environment, the rate of adaptation decreases; 2) in the absence of recombination, clonal interference dominates the adaptive process. Moreover, we will be able to infer what are the main forces shaping the adaptation of an important commensal to the mouse gut.

D21SY10PS1439

GENOMICS OF ADAPTATION DURING EXPERIMENTAL EVOLUTION OF LEGUME SYMBIONTS

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Exchange of genetic material plays a major role in bacterium evolution. Among the best illustrations are nitrogen-fixing legume symbionts that evolved and spread in many unrelated phylogenetic branches through lateral transfer of essential symbiotic genes. The full phenotypic expression of the acquired traits may require readjustment of the new genetic background. However mechanisms allowing post-LGT adaptation are largely unknown. To address this question, we took advantage of the experimental evolution of a pathogenic Ralstonia solanacearum chimera carrying the symbiotic plasmid of the rhizobium Cupriavidus taiwanensis into legume symbionts (1). The chimeric Ralstonia was progressively adapted to nodule tissues by 9 parallel lineages of serial ex planta-in planta passages. Evolution was very fast, since the two first major symbiotic steps, nodulation and intracellular infection, have been acquired in less than 16 cycles (1,2). Genome resequencing revealed an overabundance of mutations in our evolution experiment. A total of ca. 500 point-mutations were detected in the 9 final clones as compared to the original ancestor. We will present a first analysis of the genomic changes that came along with the adaptation process, i. e. mutation spectra, evolution of the number and nature of mutations and molecular convergences between lineages at the gene, operon and pathways levels.

Symposium

11. Advances in Quantitative Approaches to Recent Evolutionary Change in Humans

23 and 24 August



Program

Friday 23 August Session(s): 12, 13 Saturday 24 August

Session(s): 14

Organisers: Alexandre Courtiol and Ian Rickard

Invited speakers: Maarten Larmuseau and Stephen Stearns

Description:

Applying a rigorous evolutionary framework to understand human biology and behaviour present numerous challenges. However, many recent advances have brought this field to a point where resolution of past difficulties may be achieved. The goal of this symposium is to provide a diverse overview of the state-of-the-art in quantitative approaches to investigating human evolution at different levels of biological organisation.

D23SY11IT15:45R9

GENETIC GENEALOGY COMES OF AGE: ADVANCES IN THE USE OF DEEP-ROOTED PEDIGREES IN HUMAN EVOLUTIONARY RESEARCH

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Research on the recent human evolution will benefit from the implementation of extended genetic genealogical data. The approach to combine deep-rooted pedigrees with genetic information advances the understanding of changes in the human population genetic structure during the last centuries. This recent advance is mainly based on the extensive growth of whole genome sequencing data and available genealogical data of high quality. Moreover, according to the latest genetic genealogical research the historical non-paternity rate in Western Europe is estimated around 1% per generation within the last four centuries, which means that the expected relationship between the legal genealogy and the genetics of DNA donors exists. Therefore, genetic genealogical data will help with three research aims of human evolutionary studies: (I) detecting signals of (past) population stratification and interpreting the population structure in a more objective manner, (II) obtaining the time scale and impact of particular detected gene flow events more accurately and (III) determining temporal genetic differentiation within a population by combining in-depth pedigree data with haploid markers. Each of these research aims will be discussed with examples of the human population in Flanders (Western Europe). At the end, we will discuss the advantages and pitfalls of using genetic genealogy within studies on human evolutionary genomics.

D24SY11IT10:30R4

THE DEMOGRAPHIC TRANSITION AND THE GLOBAL BURDEN OF DEGENERATIVE DISEASE

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I advance and support the hypothesis that the demographic transition uncovered previously minor costs of reproduction that now explain much of the global burden of degenerative disease. Prior to the demographic transition, alleles were selected that improved reproductive performance early in life at the cost of vulnerability to degenerative disease in the aged, a cost that was rarely realized when most died of infectious disease or in childbirth. When mortality rates dropped during and after the transition, more people started surviving to ages where these previously evolved vulnerabilities now make a difference. Available evidence suggests that some of our vulnerability to type 2 diabetes and cancer has this antagonistically pleiotropic property. We should be looking for more such evidence, for understanding the evolutionary origins of such diseases, including cardiovascular disease and dementia, is both interesting and potentially useful. Because antagonistic pleiotropy is a property culturally mimicked by education, here again is an issue where we need better understanding of how biological and cultural evolution interact to explain current patterns and processes.

D23SY11RT16:33R9

TREES, THICKETS, OR SOMETHING IN BETWEEN? RECENT THEORETICAL AND EMPIRICAL WORK IN CULTURAL PHYLOGENY

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Anthropology has always had as one of its goals the explanation of human cultural diversity across space and through time. Over the past several decades, there has been a growing appreciation among anthropologists that the phylogenetic approaches biologists have developed to reconstruct the evolutionary relationships of species are useful tools for building and explaining patterns of human diversity. Phylogenetic methods offer a means of creating testable propositions of heritable continuity —how one thing is related to another in terms of descent. Such methods have now been applied to a number of cultural phenomena, including languages, projectile points, textiles, marital customs, and political organization. Here I discuss several cultural phylogenies and demonstrate how they were used to address long-standing anthropological issues. Even keeping in mind that phylogenetic trees are nothing more than hypotheses about evolutionary relationships, some researchers have argued that when it comes to cultural behaviors and their products, tree building is theoretically unwarranted. I examine the issues that critics raise and find that they in no way sound the death knell for cultural phylogenetic work.

D23SY11RT16:57R9

IDENTIFYING RECENT DARWINIAN SELECTION ACTING ON DIFFERENT HUMAN APOL1 VARIANTS AMONG DIVERSE AFRICAN POPULATIONS IN TRYPANOSOMIASIS ENDEMIC AREAS

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Disease susceptibility may arise as a consequence of adaptation to infectious disease. Recent findings have suggested that higher rates of chronic kidney disease (CKD) in individuals with recent African ancestry may be attributed to two *APOL1* risk alleles (G1 and G2) at the SRA-interacting domain. These two alleles appear to have risen adaptively, possibly due to their protective effects against African trypanosomiasis (African sleeping sickness). In order to explore the distribution of potential functional variation at APOL1, we studied nucleotide variation in 187 individuals across ten geographically and genetically diverse African ethnic groups with differing levels of exposure to two subspecies of *Trypanosoma brucei* that cause African sleeping sickness. We observed unusually high levels of nonsynonymous polymorphism at the functional domains that are required to lyse parasites. Whereas allele frequencies of G2 are similar across all populations (3-8%), the G1 allele is only common in the Yoruba (39%). We further identified a previously unreported haplotype (termed G3) that contains a nonsynonymous mutation at the membrane-addressing domain of APOL1 and is present in all populations except for the Yoruba. Analyses of long-range patterns of LD indicate evidence for recent selection acting on the G3 haplotype in Fulani (cattle-herding population) from Cameroon that

are endemic to Gambian trypanosomiasis, suggesting its possible protective effects against trypanosome infection. Our results indicate that the G1 and G2 variants are geographically restricted and that there are more candidate variants at other functional domains contributing to trypanosomiasis resistance and risk for CKD in African populations.

D23SY11RT17:45R9

HOW HAS THE GENETIC ARCHITECTURE OF KEY LIFE HISTORY TRAITS RESPONDED TO THE DEMOGRAPHIC TRANSITION IN A HUMAN POPULATION?

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A central issue in evolutionary biology concerns the long-term reliability of predictions of evolutionary change. Theory predicts that the genetic architecture of life history traits (summarised by the additive genetic variance-covariance matrix, G) will change over time and thus affect how traits respond to selection. However, studies have found support both for fast changes as well as for a great consistency in G over time. It thus remains unclear how rapidly and in what manner G itself changes in response to changes in selection pressures or environment. The demographic transition to low mortality and fertility rates in many recent human populations involves a drastic environmental change, but its consequences for the evolutionary potential of traits have rarely been addressed. We use genealogical data from 8 parishes in Finland, from natural high (5-6 offspring) to recent low (< 2 offspring) fertility over 350 years to address this question at the genetic level. We study four key life history traits; age at first and last reproduction, number of offspring and longevity, all of which show significant phenotypic changes during the time period. We use the animal model quantitative genetic approach to study whether and how the genetic architecture underlying these traits has also changed, by comparing the full G matrix in the periods before and after the demographic transition. First, we establish significant additive genetic variance and heritability for all traits during both time periods. Second, we present the genetic covariances and correlations between all four traits during both periods. Third, we compare the overall G matrix of the two periods to elucidate if and how G has changed during the demographic transition. The results provide a novel insight in how traits can respond to selection in contemporary human populations and whether the potential for such responses might have changed along with the recent demographic and societal changes.

D23SY11RT18:09R9

QUANTITATIVE GENETIC VARIATION, SELECTION AND SECULAR CHANGE OF SKULL SHAPE IN HUMANS

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The combined use of geometric morphometrics and quantitative genetics provides a set of powerful tools for obtaining quantitative information that is crucial for many important questions concerning the evolution of shape. In particular, the demographic information that is available for human populations make humans a unique study system for studying the mechanisms of evolutionary change in morphological traits. We investigate skull shape in the population of Hallstatt (Austria), where a collection of human skulls with associated records offer a unique opportunity for such studies. We use an individual-based statistical model to estimate the genetic covariance matrix, and characterize selection using fitness estimates from demographic data. We find clear evidence for directional selection, but not for nonlinear selection (stabilizing or disruptive selection). The predicted response to this selection, computed with genetic parameters from the population, does not match the estimate of secular change over the 150-year range of the data. We discuss possible reasons for the mismatch.

D23SY11RT18:33R9

A GENOME-WIDE SCAN FOR RELAXATION OF CONSTRAINTS IN THE HUMAN LINEAGE AFFECTING SPECIFIC FUNCTIONAL PROCESSES

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Changes in the subsistence mode of a species can lead to adaptive evolution of new functions, while it can also cause relaxed negative selection in previously essential functions. While positive selection in humans has been intensely studied, functional processes subject to relaxed constraints in the human lineage remain largely unknown. Here we present a framework for detecting relaxation of selective constraints that affect a particular functional process specifically in one taxon. Jointly using human and chimpanzee population genomic data with mammalian comparative genomic data, we identify olfactory receptors and proteasome subunits as candidates of relaxed constraints in humans: both gene sets contain high frequency non-synonymous mutations in humans while having conserved amino-acid sequences across other mammals. We further discuss the possible underlying causes of this signal.

D23SY11RT18:57R9

A QUANTITATIVE GENETICS APPROACH TO EQUILIBRIUM ASSESSMENT AND EQUILIBRIUM FITNESS ESTIMATION; AN APPLICATION TO STUDY THE POLYMORPHISM OF THE HUMAN ACP1 IN EUROPE

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Studies of equilibrium and stability under selection are liable to further development. With multiple alleles, a tiny proportion of possible genotype frequencies are possible at polymorphic equilibria. I present a quantitative genetics (QG) method for checking whether a set of genotype frequencies are equilibrium frequencies that also provides estimates of the fitnesses of all genotypes. This method is here applied to a one-locus multiallelic system, but applications to multiple-locus systems with epistasis are also possible. Method Classical OG studies focused on directional selection on the phenotype. Considering instead that the trait is fitness enables tracking genotypic frequencies under other selection regimes. Thus, if the additive variance may be minimized to zero for a set of observed frequencies, then these are putative equilibrium frequencies and the corresponding fitnesses are estimates of the equilibrium fitnesses. Two exclusive properties of NOIA are crucial for performing such analysis: it generalizes previous models and it provides explicit expressions of variance components (Álvarez-Castro and Carlborg 2007, Álvarez-Castro and Yang 2011). The human ACP1 polymorphism in Europe Three alleles of ACP1 (A, B, C) coexist in human European populations and C was claimed to be deleterious using biallelic methods. The multiallelic method proposed here shows instead that the observed genotypic frequencies are consistent with a polymorphic equilibrium with fitnesses in accordance with physiological observations (AA affected by fetal macrosomia). NOIA reveals a fast rise of additive variance for slight fitness increases of AA (not published). This may actually be leading to fixation of A in European populations since fetal macrosomia--with the aid of pregnancy monitoring and reasonably safe caesarean sections--entails nowadays less of a problem than it did some decades ago. ALVAREZ-CASTRO & CARLBORG 2007 Genetics 176 ALVAREZ-CASTRO & YANG 2011 Genetica 139

D24SY11RT11:18R4

UPDATING THE Y-CHROMOSOMAL PHYLOGENY BASED ON SNPS IN AVAILABLE WGS DATA

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The sex-determining Y-chromosome is an interesting marker for many human evolutionary and population genetic studies, as it accurately tracks patrilineal descent. For these studies a high-resolution state-of-the-art Y-chromosomal phylogeny is essential to characterize Y-chromosomes. Due to the rapid progress in next generation sequencing technologies, an explosion of human whole genome sequencing (WGS) data has become available. WGS data is useful to verify, optimize and extend the current phylogeny for haploid markers such as single nucleotide polymorphisms in the Y-chromosome (Y-SNPs). We present two new open access software packages which analyse WGS data for Ychromosomal phylogenetic applications. First, the AMY-tree software determines the haplogroup for a sample based on the Y-SNPs derived from WGS experiments and it also indicates ambiguities such as recurrent SNPs within the used tree. Because of these features AMY-tree is able to include all reported Y-SNPs within one phylogenetic framework. Second, the PENNY software detects new informative Y-SNPs from WGS data by in silico analysis to increase the resolution of the Y-chromosomal tree. The AMY-tree and PENNY software and all their capabilities were tested in detail on more than 750 male full genomes with different geographical origins. An analysis of these data provides a much more detailed tree than the last 'officially' published tree of the Y Chromosome Consortium (YCC) in 2008. On the other hand, further sequencing will still be required since currently full genomes of only a limited set of Y-chromosomal (sub-)haplogroups are available. Overall, these advances of the Ychromosomal tree will provide many applications for research on recent human evolution.

D24SY11RT11:42R4

SELECTION ON PENIS SIZE, BODY SHAPE AND HEIGHT IN HUMANS: A SIMPLE MULTIVARIATE METHOD TO QUANTIFY FEMALE PREFERENCES BASED ON MALE PHYSICAL ATTRACTIVENESS

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Compelling evidence from many animal taxa indicates that male genitalia are often under postcopulatory sexual selection for characteristics that increase a male's relative fertilization success under sperm competition. There could, however, also be direct pre-copulatory female mate choice based on male genital traits. Before clothing, the non-retractable human penis would have been conspicuous to potential mates. This, in combination with claims that humans have a large penis for their body size compared to other primates, has generated suggestions that human penis size partly evolved due to female choice. We presented women with digitally projected fully life-size, computer-generated animations of male figures to quantify the (interactive) effects of penis size, body shape and height on female assessment of male sexual attractiveness. We generated 343 male figures that each had one of seven possible values for each of the three test traits (7x7x7 = 343). All seven test values per trait were within two standard deviations of the mean based on a representative sample of males. We calculate response (fitness) surfaces based on the average attractiveness rank each of the 343 male figure received. We also calculated individual response surfaces for 105 women (each women viewed 53 figures). Both methods yielded almost identical results. We discuss our finding in the context of previous studies that have taken a univariate approach to quantify female preferences. We discuss the hypothesis that pre-copulatory sexual selection might play a role in the evolution of genital traits.

POSTERS

D23SY11PS0257

GENETIC CONSTRAINTS UNDERLYING HUMAN REPRODUCTIVE TIMING IN A PRE-MODERN SWISS VILLAGE

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The trade-off between reproductive investment in early versus late life is central to life-history theory. Despite abundant empirical evidence in support of different versions of this trade-off, the specific trade-off between age at first reproduction (AFR) and age at last reproduction (ALR) has received little attention, especially in long-lived species with a pronounced reproductive senescence such as humans. Using genealogical data for a 19th-century Swiss village, we (i) quantify natural selection on reproductive timing, (ii) estimate additive genetic (co)variances, and (iii) use these to predict evolutionary responses. Selection gradients were computed using multiple linear regressions, and the additive genetic variance-covariance matrix was estimated using a restricted maximum-likelihood animal model. We found strong selection for both an early AFR and a late ALR, which resulted from selection for an earlier and longer reproductive period (RP, i.e. ALR-AFR). Furthermore, postponing AFR shortened RP in both sexes, but twice as much in women. Finally, AFR and ALR were strongly and positively genetically correlated, which led to a considerable reduction in the predicted responses to selection, or even rendered them maladaptive. These results provide evidence for strong genetic constraints underlying reproductive timing in humans, which may have contributed to the evolution of menopause.

D23SY11PS0735

FITNESS MEETS FITNESS: TAKING AN EVOLUTIONARY APPROACH TO PHYSICAL FITNESS IN HUMANS

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Evolutionary biologists are usually the first to emphasize that Darwinian fitness has little in common with the meaning that is commonly attached to fitness by the general public, that is physical fitness. However, recent studies in humans and non-human animals are suggestive of an important role for physical fitness in shaping variation in Darwinian fitness via natural and sexual selection, both in the past and in the present. Indeed, it has been argued that it is selection on physical performance that has made us who we are today. To gain a better understanding of variation in (physical) fitness, I use concepts and methods from evolutionary biology and life-history theory and apply these to data on human running performance. Specifically, using a large (longitudinal and cross-sectional) data set for running performance by both men and women over a wide range of distances, I test for sex differences, the effects of ageing and training, and for trade-offs between long- versus short-distance and generalists versus specialists. I will use these findings to infer the selective pressures acting on physical fitness in humans, and to argue that Darwinian and physical fitness may have more in common with each other than is often assumed.

D23SY11PS0747

EXPERIMENTAL STUDIES OF HUMAN SOCIAL LEARNING STRATEGIES: EXPLORING SEX DIFFERENCES

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Objectives Culture is an important driver of recent biological evolution in humans. The mechanisms by which information is transmitted between individuals can be studied at the population level - by cultural evolutionists, and at the individual level – by social psychologists. We combined methods from these two approaches to investigate how sex differences in confidence might lead to sex differences in the use of a copy-when-uncertain social learning strategy. Methods Participants (Study 1: N=97; Study 2: N=89) completed a series of two-alternative forced-choice puzzles and reported their confidence in each answer. They then saw the decisions of some previous participants before being asked again for their answer. Social information use was inferred when participants switched their answer to match that of the majority. We modelled the probability of social information use with participant sex, confidence in initial decision, and accuracy of initial decision as predictors. Results Across both studies, confidence had a large effect on social information use, indicative of a copy-when-uncertain strategy. Accuracy predicted confidence, indicating that this strategy is adaptive. Confidence also differed by sex: women reported lower confidence (independent of any small sex differences in accuracy), which in turn increased their probability of using social information. Conclusions Although both sexes appear to use a 'copy-when-uncertain' strategy, women are more likely to feel uncertain. This means that a strategy observed to be used in a population (e.g. copy-when-uncertain) can vary according to individual differences in psychological traits. Further integration of these two levels of explanation is therefore needed.

D23SY11PS0802

EVOLUTION OF A CO-EXPRESSION NETWORK INVOLVED IN PRIMATE BRAIN FUNCTIONS: CLUES TO THE EVOLUTION OF HUMAN SPECIFIC COGNITIVE ABILITIES

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Cognitive abilities are among the most striking differences between humans and other primates. As cognition is a complex trait it is beneficial to analyze its evolution on multiple levels. Here we analyzed genome wide expression data of several human, chimpanzee, and rhesus macaque brains to identify molecular changes that may underlie the evolution of human-specific cognitive skills. Because transcription factors (TFs) are main players in regulating gene expression, we focused on TF changes that might drive expression pattern differences between the species. For each TF with a species-specific expression level we identified co-expressed genes, which are potential target genes or interaction partners of the respective TFs. Using this information we derived weighted topological overlap (wTO) networks in which the nodes are the TFs and the weighted links represent the commonality of the TFs' co-expressed genes. We revealed: 1. The ancestral network, consisting only of links common to all three species, was involved in cognitive functions, such as forebrain development, synaptic plasticity, and learning, as inferred from the Gene Ontology (GO) terms enriched among the genes co-expressed with the most TFs in this network. 2. In the human network, TFs are more interconnected than in the chimpanzee and rhesus macaque network, indicating more complex regulation of genes involved in cognition in the human brain. 3. The TF sub-network constructed only from genes with a humanspecific expression change was enriched for GO terms that point to changes in energy metabolism, learning and memory processes in the human brain. Moreover, only in this sub-network, but not in the sub-networks of the other species, we identified an excess of TFs implicated in mental disorders. Taken together, in a network of TFs that is predicted to regulate cognitive functions we identified evolutionary changes that are potentially involved in the evolution of human- specific cognitive abilities.

D23SY11PS0905

MULTIPLE INSTANCES OF ANCIENT BALANCING SELECTION SHARED BETWEEN HUMANS AND CHIMPANZEES

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Balancing selection, in which two or more alleles are maintained in a population by selection, is predicted to lead to high diversity and to haplotypes with deep coalescence times. Moreover, if selection pressures are older than species split times, species may share alleles identically by descent. Such balanced polymorphisms are thought to be extremely rare, with a few notable examples involved in host immunity, such as at the MHC in primates and R genes in plants. However, modeling suggests that the footprint of balancing selection may be difficult to detect. Using genome-wide sequence data for 10 Western chimpanzees from the PanMap project and 1000 Genomes Pilot 1 data, we undertook a genome-wide search for orthologous sites polymorphic for the same alleles in both chimpanzee and human. We found that SNPs are shared in excess of what is expected by chance after accounting for local variation in the mutation rate. While it is difficult to distinguish balanced polymorphism from recurrent mutation for a single SNP, the short ancestral segments on which a balanced polymorphism resides may contain additional shared ancestral polymorphisms. We therefore focused on cases of two or more shared SNPs in close proximity and with the same LD patterns in both species, a scenario that is unlikely to occur by recurrent mutation. Besides the MHC, we identified 125 candidate loci, only

two of which overlap exons. Notably, nearby genes are enriched for membrane glycoproteins, which are often found at host-pathogen interfaces. For five of the loci, more than two pairs of SNPs are shared with the same LD pattern and a phylogenetic tree clusters by haplotype rather than by species, providing strong evidence that the polymorphisms are ancestral and pointing to new targets of selection. These results suggest that balancing selection has acted on regulatory variation in both humans and chimpanzees and that pathogens may be a common pressure leading to such long-term balancing selection.

D23SY11PS1097

THE DEMOGRAPHIC TRANSITION INFLUENCES VARIANCE IN FITNESS AND SELECTION ON HEIGHT AND BMI IN RURAL GAMBIA

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Recent human history is marked by demographic transitions characterized by declines in mortality and fertility. By influencing the variance in those fitness components, demographic transitions can affect selection on other traits. Parallel to changes in selection triggered by demography per se, relationships between fitness and anthropometric traits are also expected to change due to modification of the environment. Here we explore for the first time these two main evolutionary consequences of demographic transitions using a unique data set containing survival, fertility, and anthropometric data for thousands of women in rural Gambia from 1956–2010. We show how the demographic transition influenced directional selection on height and body mass index (BMI). We observed a change in selection for both traits mediated by variation in fertility: selection initially favored short females with high BMI values but shifted across the demographic transition to favor tall females with low BMI values. We demonstrate that these differences resulted both from changes in fitness variance that shape the strength of selection and from shifts in selective pressures triggered by environmental changes. These results suggest that demographic and environmental trends encountered by current human populations worldwide are likely to modify, but not stop, natural selection in humans.

D23SY11PS1202

WHY IS CHILDBIRTH SO HARD IN HUMANS?

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Compared to other primates, childbirth is remarkably difficult in humans, the reason being that the head of the human fetus is large in comparison to the birth-relevant dimensions of the human pelvis. Modern human pelvic morphology must serve more than one purpose: It is thought to have evolved as a compromise between being shaped for upright walking and giving birth to large-headed neonates. This "obstetric dilemma" arose as a consequence of bipedal humans evolving increasingly larger brains. Although anatomically modern humans have existed for at least 100,000 years, and although the selection pressure for evolving wider birth canals has probably been considerable throughout, we do not see any evolutionary response in birth-relevant pelvic dimensions. We hypothesize that this lack of response is due to evolutionary constraints that inhibit the evolution of wider birth canals in humans. In this study we try to identify allometric constraints using morphometric techniques on a human skeletal sample.

D23SY11PS1226

INVESTIGATING THE CO-EVOLUTION BETWEEN HUMAN LACTASE GENE AND DAIRY CULTURE IN EUROPE

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Genes that have undergone recent positive selection during human evolution can provide new insights into the evolutionary forces that have shaped our species. Human *lactase* gene *LCT* which encodes the enzyme responsible for hydrolysis of lactose, the milk sugar is under recent positive selection. The production of lactase is declined after the weaning period; however, in some individuals, lactase expression persists into adulthood. Lactase persistence is found at high frequency in northern and western European populations. Based on Principal Component Analysis (PCA) of genetic diversity of six markers in cattle milk protein genes, Beja-Pereira et al. (2003) proposed that since Neolithic times, there has been co-evolution between LCT in humans and cattle milk protein genes. To test this hypothesis, we used dense SNP datasets for human (441,845 SNPs) and cattle (44,706 SNPs) populations to investigate the signatures of positive selection at *LCT* and cattle milk protein genes in Europe. To this end, we used a new genome-scan method based on a diffusion approximation for the distribution of allele frequency at migration-selection-drift equilibrium (Vitalis et al, in prep). For *LCT*, we detected a north-south gradient for the intensity of positive selection. For the cattle data, we could not detect any signature of positive selection in cattle milk protein genes. Furthermore, PCA was performed using the markers surrounding cattle milk protein genes provided the same pattern of geographical variation as with the complete cattle genomic data, showing a north-south gradient of variation across Europe. The results of our study therefore provide no evidence for the co-evolution of lactase and cattle milk protein genes. We argue that the coincidence between the gradient in the selection intensity at *LCT* in humans and geographic variation in cattle genes simply reflect the migration history of both species during the Neolithic along the Danubian route.

THE VALUE OF REPUTATION

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Reputation plays an important role in fostering human cooperation. A distinctive feature of reputationbased mechanisms is their capability to maintain cooperation among strangers engaged in one-shot interactions. In evolutionary biology, reputation provides a fascinating alternative to direct reciprocity, group selection or interactions between relatives for explaining the evolution of altruistic behavior. In the social sciences, reputation is of interest because one-shot interactions in global and electronic markets are more and more replacing traditional, repeated interactions within long-lasting communities. Empirical and theoretical work from both fields indicates that a good reputation is valuable in that it increases one's expected payoff in future interactions. Here we present laboratory experiments for quantifying the value that participants place on a good reputation. To do so, we couple a Prisoner's Dilemma (PD) game where participants can earn a good reputation with a market where reputation can be bought and sold. We show that the more valuable a good reputation is in the PD game, the higher are the prices at which it is traded in the market, suggesting that participants can assess the value of a good reputation. We also find that in the presence of trading, cooperation is maintained at a lower level then in the absence of trading. This suggests that trading of reputation can have a detrimental effect on cooperation, perhaps through "crowding out" of the participants' intrinsic motivation to cooperate. However, our experiments also demonstrate that trading of reputation can promote cooperation. When participants play a short fixed-length series of Prisoner's Dilemmas, the opportunity to sell good reputation when exiting the game can mitigate end-game effects. Thus reputation markets could potentially create value and increase social welfare when applied in an appropriate context.

Symposium

12. Studying Multigene-Family Evolution in the Post-Genomic Era: Towards a Population Genomic Approach to MHC Evolution

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3

Organisers: Reto Burri and Robert Ekblom

Invited speakers: Jim Kaufman, Jacek Radwan and David

Richardson

Description:

Multigene families, like the MHC, encode central components in pathways involved in individual fitness and are key to important adaptations. Elucidating the molecular processes involved in their evolution is an important issue to further our understanding of the genetic basis of biological diversification. Here, we aim at bringing together researchers studying the functional genomics, evolutionary ecology and population genomics of MHC to discuss the most recent developments in each of the fields and develop new ideas and future research directions that integrate the different branches of MHC evolution in particular and multigene family evolution in general.

D20SY12IT10:30R9

GENE CO-EVOLUTION IN ACTION: HOW GENOMIC ORGANISATION IMPACTS EVOLUTION OF THE ADAPTIVE IMMUNE SYSTEM OF JAWED VERTEBRATES

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A crucial part of the adaptive immune system of jawed vertebrates is recognition by T lymphocytes of pathogen components presented by molecules of the major histocompatibility complex (MHC). MHC genes often have high levels of allelic polymorphism and sequence diversity, thought to be due to an on-going arms race with pathogens. However, the strong genetic associations of humans are with autoimmune disease. Starting with the chicken MHC as a model, we have found that key features of the MHC genomic organisation and subsequent function differ between typical mammals and many (if not most) non-mammalian vertebrates. In particular, the chicken MHC is organised differently than typical mammals, which leads to haplotypes of polymorphic interacting genes, which in turn leads to single dominantly-expressed MHC class I and class II molecules, which finally leads to strong genetic associations with resistance and susceptibility to infectious pathogens. The salient features discovered for the chicken MHC are shared with many non-mammalian vertebrates and are likely ancestral, with the organisation of the typical mammalian MHC arising from an inversion. However, among jawed vertebrates there are at least two other functional strategies for the MHC which have arisen independently, and still others (like passerine birds) not well understood. The co-evolution between antigen processing and antigen presenting genes should mean that evolution is slower in chickens than in mammals, so we have tried to understand the molecular basis for function in an historic recombinant, and have screened many commercial chickens and other populations for such recombinants.

D20SY12IT14:00R9

WHAT DRIVES THE RAPID REGENERATION OF MHC DIVERSITY AMONGST RECENTLY BOTTLENECKED POPULATIONS?

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.Population bottlenecks can restrict variation at functional genes, reducing the ability of populations to adapt to new and changing environments. Understanding how populations generate adaptive genetic variation following bottlenecks is therefore central to evolutionary biology. The major histocompatibility complex (MHC) is a multigene family that provides an ideal model for studying adaptive genetic variation due to its central role in pathogen recognition. While de novo MHC sequence variation is generated by point mutation, gene conversion can generate new haplotypes by transferring sections of DNA within and across duplicated MHC loci. However, the extent to which gene conversion generates new MHC haplotypes in wild populations is poorly understood. We used a 454 sequencing protocol to screen MHC variation across all 13 island populations of Berthelot's pipit (Anthus berthelotii). The recent colonisation of this species (<75,000 years ago), along with the replicated island system gave us a unique opportunity to identify which MHC alleles were involved in the original colonisation events, and which have been generated in situ, post colonisation. This, in turn, allowed us to identify how new MHC haplotypes have been regenerated across the island populations. In light of these results, I will discuss the roles of mutation, gene conversion and selection in generating and maintaining functional genetic diversity in bottlenecked populations.

D20SY12IT14:48R9

EVOLUTION OF MHC GENE NUMBER: OPTIMALITY HYPOTHESIS AND BEYOND

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MHC genes code for proteins involved in recognition of pathogens. Their extreme polymorphism, thought to be driven primarily by selection from parasites, has been subject of much theoretical and empirical work. In addition to allelic diversity, MHC genes are often multiplicated, but the evolution of the MHC copy number received relatively less attention. Optimality hypothesis (Nowak et al. 1992) poses that an increase in parasite recognition capabilities with increased number of MHC molecules expressed is traded off against higher rate of deletion of auto-reactive lymphocytes, thus some intermediate individual MHC diversity should be favoured. Empirical tests of this hypothesis have been hampered by technical difficulties associated with the typing of multi-locus genotypes, but this has now been overcome by new generation sequencing methods. I will review the recent empirical studies of the optimality hypothesis. The presence of multiple MHC genes in the genome may also facilitate creation of new alleles via inter-genic recombination (Ohta 1991). The results of simulations of host-parasite coevolution showed that such newly created alleles are very likely to be retained in populations. This opens the possibility for haplotypes with a high number of MHC copies to hitch-hike with positively selected alleles.

D20SY12RT11:18R9

IN SILICO PEPTIDE-BINDING PREDICTIONS OF MHC CLASS I REVEAL SIMILARITIES ACROSS DISTANTLY RELATED SONGBIRD SPECIES, SUGGESTING CONVERGENCE ON THE LEVEL OF PROTEIN FUNCTION

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Many songbirds have a large number of transcribed MHC class I genes compared to most mammals. To elucidate the reasons for this large number of genes and to search for non-classical MHC genes in songbirds, we compared songbird MHC class I alleles (α 1- α 3 domains). By applying phylogenetic analysis, homology modelling and in silico peptide-binding predictions, we could compare both functional and genetic relationships among transcribed genes. We found more pronounced clustering of the MHC class I allomorphs (allele-specific proteins) in regard to their protein function (peptidebinding specificities) compared to their genetic relationships (amino acid sequences), indicating that the high number of alleles is of functional significance. The MHC class I allomorphs from house sparrow and tree sparrow, species that diverged ten MYA, had overlapping peptide-binding specificities. These similarities across species were also confirmed in phylogenetic analyses, and were similar for genes that we interpret as classical and non-classical. Remarkably, there were also overlapping peptide-binding specificities in the allomorphs from house sparrows and great reed warblers, although these species diverged about 30 MYA. This overlap was not found in a tree based on amino acid sequences. Our interpretation is that convergent evolution on the level of the protein function, possibly driven by selection from shared pathogens, has resulted in allomorphs with similar peptide-binding repertoires, although trans-species evolution in combination with gene conversion cannot be excluded.

D20SY12RT11:42R9

SEXUAL SELECTION MORE THAN PARASITISM EXPLAINS FUNCTIONAL VARIATION OF THE MHC ACROSS MAMMALS

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Diversity at the major histocompatibility complex (MHC), vitally important for vertebrate immune defense, varies widely across species. Parasites have been identified as a major evolutionary force driving MHC polymorphisms across species, but sexual selection and disassortative mating is another likely mechanism. MHC-based mating preferences have been observed for multiple species including humans, but the generality of mate choice as a driver of MHC polymorphism in the wild is debated. In reality, both parasite-mediated selection and sexual selection may act in concert in wild populations. To investigate potential contributions of parasitism and sexual selection in explaining among-species variation in MHC diversity, we used comparative methods to examine measures of MHC diversity across 115 mammal species, including carnivores, chiroptera, primates, rodents, and ungulates. Specifically, we tested whether parasite species richness and relative testes size (as an indicator of sexual selection) were correlated with two measures of MHC class II DRB diversity: allelic richness and nucleotide diversity. Controlling for mammal phylogeny, neutral mutation rate and confounding ecological variables (i.e. population size, body mass, and sampling effort), we found that parasite species richness was positively correlated with MHC nucleotide diversity for bats and ungulates, and negatively correlated for carnivores. In contrast, relative testes size was positively correlated with MHC nucleotide diversity for carnivores, rodents, and ungulates, and for all taxa combined. Mammal taxonomic group was the strongest predictor of MHC allelic richness, with ungulates having lower diversity in general. This study provides support for both parasite-mediated selection and sexual selection in shaping variation in functional MHC polymorphism across a broad suite of mammals, and importantly, suggests that sexual selection may be more ubiquitous than previously thought.

D20SY12RT15:45R9

INTERPLAY OF GENE CONVERSION AND CROSSOVER IN THE MOLECULAR EVOLUTION OF MULTIGENE FAMILIES

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Multigene families are part of the pervasive Structural Variation (SV) present in eukaryotic genomes. In humans, regions with SV have been associated to disease and have been shown to participate in evolutionary innovation. Despite its widespread abundance and functional relevance, an accurate description of the underlying forces shaping the evolution of these regions is still lacking. In particular, the proper characterization of the interplay between mutation, crossover and gene conversion in multigene families is fundamental. We have developed a forward-time simulation program that incorporates duplications and focuses on the effect of concerted evolution (the non-independent evolution of duplicated regions). By means of simulations, we have explored a wide range of parameters, gaining insight into the evolution of regions under concerted evolution such as the MHC. First, we observe that neutral concerted evolution can confound scans for selection by mimicking the effects of both weak purifying selection or weak positive selection. These effects can be more pronounced if duplicated regions are collapsed, as is frequent in low-quality genome assemblies. Additionally, we explore the effects of crossover hotspots in duplicated regions that present gene conversion activity. Our results show that multigene-family evolution is highly dependent on the spatial distribution of crossover and gene conversion events and their rates, even under neutrality.

D20SY12RT16:09R9

LINKING CHEMOSENSORY MULTIGENE-FAMILY EVOLUTION WITH SPECIATION IN THE PEA APHID (ACYRTHOSIPHON PISUM)

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Ludovic DUvaux, Carole Smadja, Julia Ferrari, Within and between species, multigene families (MF) are known to be highly diverse in terms of both Copy Number Variation (CNV) and allelic diversity. Accordingly, within and between population diversities of MF have a strong potential to result from, or to fuel, local adaptation - and possibly speciation. However, these diversities have seldom been contrasted and their relative evolutionary dynamics remain poorly understood. The host-plant races of the pea aphid provide an excellent system to understand these dynamics in the context of speciation with gene flow. Notably, Chemosensory MF (CMF) - e.g. Olfactory Receptors (OR), Gustatory Receptors (GR), Odorant Binding Proteins (OBP) - are critically important for specific host plant recognition, i.e. putatively a main cause of reproductive isolation. We sequenced 120 individuals from 8 races using a target enrichment protocol and Solexa sequencing – guaranteeing a median coverage of 150X. In doing so, we assessed CNV and nucleotidic diversity at about 3000 exons (from CMF, other MF and control genes) and 650 promoters of CMF genes. Preliminary results show CNV is widespread, as it occurs in 65% of all exons (even surprisingly up to 57% for control exons). As with SNPs in single copy genes, most CNV in non CMF genes is shared among races. In contrast, for targets linked to chemosensory genes – notably GR exons and promoters – CNV tends to structure by race. Also, the rate of duplication appears higher for CMF: OR, OBP and promoters show significantly more CNV than other MF. Together, these results suggest that many CMF genes may evolve under positive selection and contribute to adaptation to host plants. In order to better understand the genetic basis of adaptation, work is ongoing to link patterns of CNV (i) to the history of gene flow between races; and (ii) to observed differences in gene expression within and between races across native and non-native host plants.

D20SY12RT16:33R9

EVOLUTION OF OLFACTION IN TUBENOSE SEABIRDS

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The sense of olfaction is one of the most ancient senses with which vertebrates collect information about their surroundings, playing a critical role in survival and reproduction. We are extending the understanding of the evolution of the olfactory receptor (OR) genes in the avian group, by focusing on Procellariiform seabirds. This group has one of the largest relative olfactory bulb sizes among extant birds, and other anatomical features correlated with high olfactory capability. We built a cosmid library of Cory's Shearwater (Calonectris diomedea) gDNA, which was screened with partial shearwater OR genes. 96 positive clones were sequenced with a combination of Sanger (cosmid ends), Illumina and 454 sequencing technologies. The resulting hybrid assembly has a cumulative length of 7.4Mb, including 399 unique scaffolds (2.6Mb), and ~ 21K degenerate scaffolds. A total of 144 ORFs (> 75 amino acids) have a significant similarity to OR genes, as determined with HMMs built from bird OR genes. Many are full length OR genes, and provide the basis for a comprehensive study of OR evolution in a lineage that relies extensively on olfaction. Preliminary phylogenetic analyses of shearwater, chicken, zebra finch and lizard OR genes revealed at least three distinct clades in Cory's Shearwater. A few genes clustered with those of the remaining species in a clade, *gamma*, that predates the divergence of birds. However, most of the sampled shearwater genes belong to the avian-specific *qamma*-c clade, within which sequences cluster by species, consistent with a scenario of ubiquitous, lineage-specific expansions and/or the action of concerted evolution. Ongoing analyses will address the evolutionary history and genomic context of OR genes in this avian clade, namely the possible genomic association with MHC genes, as well as sequence diversity within and between clades, in particular in putative ligand-binding pockets encoded by shearwater-specific OR lineages.

D20SY12RT16:57R9

GENETIC SIGNATURE OF LOCAL ADAPTATION OF MHC GENES TO VARYING PARASITE PRESSURES

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Although crucial for the understanding of adaptive evolution, genetically resolved examples of local adaptation are rare. To maximize survival and reproduction in their local environment, hosts should resist their local parasites and pathogens. The major histocompatibility complex (MHC) with its key function in parasite resistance represents an ideal candidate to investigate parasite-mediated local adaptation. Using replicated field mesocosms, stocked with second-generation lab-bred three-spined stickleback hybrids of a lake and a river population, we show local adaptation of MHC genotypes to population-specific parasites, independently of the genetic background. Furthermore, the multi-locus specificity of MHC genes revealed that increased individual allele divergence of lake MHC genotypes allows lake fish to fight the broad range of lake parasites, whereas more specific river genotypes confer selective advantages against the less diverse river parasites. Such results demonstrate that not only the presence of given alleles is important for local adaptation but also that the combinations of alleles on the different duplicated loci are crucial for host-parasite coevolution.

POSTERS

D20SY12PS0391

MATE CHOICE, MULTIPLE MATING AND MHC VARIATION IN SEA TROUT (SALMO TRUTTA)

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Understanding the factors that affect spatio-temporal levels and distribution of genetic diversity is essential for the management of genetic resources in natural populations. Traditionally, neutral markers have been utilised to examine the extent of migration-drift equilibrium within populations, but this ignores how sexual selection and plasticity in life strategy can influence patterns of adaptive genetic diversity that may directly influence individual fitness and population viability. Here we examine how MHC variation is maintained by mate choice decisions and multiple paternity in a population of sea trout in NE Scotland. Parentage of 1519 S. trutta offspring from 72 nests was characterised using ten microsatellite loci and one MHC-linked minisatellite locus. The level of multiple paternity was examined within and among nests and both sexes were found to practice multiple mating, with females mating with up to 11 males per nest and males with up to eight females. The level of amino acid dissimilarity among MHC alleles between consort and non-consort males was assessed and the overall levels of MHC diversity within nests under different mating scenarios was quantified to examine how extra pair paternity influences overall brood diversity. Multiple paternity is likely to increase genetic variation in a female's offspring therefore maximising the chances that some offspring will be genetically equipped to deal with selective pressures. This bet hedging strategy may be pronounced in species, such as salmonids, which produce large broods, have high mortality rates in their first year, and spawn in their natal breeding grounds, which may heighten the risk of inbreeding. Mate choice towards consort males may enhance the chances of producing viable offspring as a more direct mechanism of estimating 'good genes'.

D20SY12PS0392

PHYLOGENETIC ORIGIN OF THE AVIAN MHC CLASS IIB

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The major histocompatibility complex (MHC) multigene family encodes genes involved in the vertebrates' adaptive immune response. The exceptional evolutionary dynamics of MHC genes, including high rates of gene duplication and recombination, qualifies them as an ideal system to understand multigene family evolution. Mammalian MHCIIB paralogs evolve independently and their duplication history can commonly be traced back over tens of millions of years. Recent studies on birds report also strong evidence for the persistence of two ancient MHC class IIB lineages over at least 100 million years. At odds with this result, only few species studied so far appear to have retained both MHCIIB lineages, and it is unclear whether they have gone largely undetected, or whether they have been lost at high frequency. In the present study we therefore isolated MHCIIB genes from species all over the avian phylogeny. We aim at reconstructing the phylogenetic history of avian MHCIIB in order to provide an accurate estimate for the time of origin of the two ancestral avian MHCIIB lineages and estimate the rates of gene loss in each lineage. Qualitative results show that the two avian MHCIIB lineages evolved prior to the evolution of extant birds, and suggest the action of generally high, but strongly variable and phylogenetically unconstrained rates of concerted evolution in the avian MHCIIB. Thereby the present study provides important insights into the avian MHCIIB long-term evolutionary history and thus provides new information in unprecedented details about the evolution of an enigmatic multigene family.

D20SY12PS0401

MHC EVOLUTIONARY ECOLOGY IN EUROPEAN BARN OWLS (TYTO ALBA)

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Major histocompatibility complex (MHC) genes play an essential role in the adaptive immune response and thus constitute a good model to study adaptive genetic variability. The extraordinary diversity exhibited by MHC genes is thought to be maintained by pathogen-driven selection, for which three hypotheses have been proposed: rare-allele advantage, heterozygote advantage and fluctuating selection. Despite a growing amount of empirical data, it is still difficult to distinguish the relative role of the different forms of selection in maintaining MHC variability. Moreover other mechanisms such as sexual selection through mate choice can shape the MHC diversity. The barn owl (*Tyto alba*) exhibits a number of characteristics that make it an ideal model to study MHC diversity. Indeed it is one of the most widespread birds worldwide and it has been shown that this species exhibits mate choice related to an ornamental trait that is associated to parasite resistance. In addition, this bird possesses a relatively simple MHC organization, and is one of only few species in which the two functional MHC class IIB genes can be amplified specifically. Using 454 technology, we sequenced exon 2 of the two MHC class IIB loci in a large number of individuals sampled throughout Europe (N=384) and Switzerland (N=960), for the latter of which information related to individual fitness is available (e.g. immunocompetence, parasite fecundity). We examined the effects of particular alleles and MHC diversity on these fitness parameters. First results indicate a positive effect of heterozygosity on parasite resistance, in agreement with expectations of the heterozygote advantage hypothesis. With the sampling across Europe we will be able to study spatial patterns of MHC diversity and evaluate if MHC genotypes are locally adapted or under balancing selection at the spatial scale. Therefore, this study provides interesting perspectives for the understanding of MHC evolutionary ecology.

D20SY12PS0408

TOLL-LIKE RECEPTOR (TLR) EVOLUTION IN WILD VERTEBRATES

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Members of the Toll-like receptor (TLR) gene family play an important role in pathogen recognition and the initiation of innate and adaptive immune responses. However, their role in host-parasite interactions and adaptation has rarely been considered in an eco-evolutionary context, and little is known about the selective pressures that have shaped TLR evolution across species and populations. Using an integrative approach that combines analyses of molecular evolution across several rodent species, large scale analyses of diversity and differentiation across populations, and ecological research on within-population associations between TLR polymorphisms and parasite susceptibility, I show that TLRs are evolutionarily dynamic and a target of parasite-mediated selection in wild vertebrates. The findings of these studies contribute to a better understanding of evolutionary processes in multigene families other than the MHC, and the selective pressures that contribute to the maintenance of immunogenetic diversity in the wild.

D20SY12PS0475

GENETIC STRUCTURE, SELECTION AND DUPLICATION IN EUROPEAN EELS MHC CLASS II GENES

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Understanding adaptation and selection in populations facing environmental changes is a major concern. Our project aims to understand the adaptation and resilience of endangered catadromous European eel to increasing parasitic pressure. Anguilla anguilla has encountered successive waves of exotic invasive parasites, namely, the swimbladder nematode Anguillicoloides crassus. Infested individuals have high swimming performance costs; and might fail to complete the reproductive migration back to the Sargasso Sea. Prevalence of such parasites is much higher in the naive European populations compared to native Japanese hosts. However, the nature of the ongoing adaptation and impact on eel's biology remain largely unknown. Given the role of Major Histocompatibility Complex (MHC) genes in host–parasite interactions, we designed specific primers to sequence 2 class II exons. Using NGS, we sampled alleles from 12 European populations (N = 120) collected throughout the species distribution range, as well as from one American population (N=10). Additional historical samples (1874) from Charente were included. Our results indicate success in isolating relevant MHC class II genes; we identified respectively 149 and 154 DAA and DAB MHC class II alleles. The multiple copy nature of the MHC loci was confirmed, with at least 4 and 5 gene copies for DAA and DAB loci, respectively. European and American eels were found to share the same allelic pool, a result in sharp contrast with microsatellite data, suggesting that balancing selection is acting on these genes while ancient polymorphisms is retained within the Atlantic lineages. There was no evidence for temporally fluctuating selection pressure. Finally, the eel MHC class II exons appeared to be under positive selection and at the upper range of rho and omega values in teleosts. These results suggest that recent duplications occurred in the genus that might have facilitated inter-locus recombination and high allelic diversity.

D20SY12PS0524

MHC EVOLUTION IN NEOTROPICAL CICHLIDS (AMPHILOPHUS SP.)

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The Major Histocompatibility Complex (MHC) is a key component of the adaptive immune system of all vertebrates. Considerable evidence has accumulated suggesting that parasite-mediated selection acting on MHC genes may ultimately lead to speciation. The Midas cichlid species complex of the crater lakes of Nicaragua is an ideal model system for the study of speciation, because it has recently colonized several isolated crater lakes where it has independently diverged into sympatric sister species. We evaluate the hypothesis that host parasite interactions might be contributing to the speciation process in this system. To this end we have characterized and sequenced the MHC class IIB of individuals from different putative species of this cichlid species complex.

D20SY12PS0933

NEXT GENERATION SEQUENCING REVEALS HIGH COMPLEXITY OF THE MAJOR HISTOCOMPABILITY COMPLEX CLASS I EXON 3 IN SEDGE WARBLER

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The major Histocompability Complex (MHC) class I genes have received a considerable of interest from evolutionary biologists. Their role in parasite recognition and extreme polymorphism makes these genes a paradigm for studying genetic effects on survival, mate choice and pathogen resistance. Exon 3 of MHC class I genes encodes a part of the domain that binds and presents peptides from pathogens. Species possessing many highly polymporphic MHC loci are potentially under strong natural selection from parasites. Determining selection acting on MHC in natural population of such species over extended periods of time offers a great opportunity to reveal mechanisms maintaining MHC diversity. Here we present initial characterization of MHC class I genes diversity in a sedge warbler Acrocephalus schoenobaenus from population inhabiting natural wetlands in the Nida valley, SE Poland. We designed primers and determined diversity at MHC class I exon 3 both in cDNA and gDNA of 4 unrelated individuals with use of Next Generation Sequencing methods. We found extreme diversity at the MHC class I exon 3 of the sedge warbler. This data will be used for designing specific primers amplifying functional alleles of MHC class I exon 3 in sedge warbler. Subsequently, we are going to infer long-term selection on MHC class I in the species.

D20SY12PS0939

EVOLUTION OF TWO MULTIGENE FAMILIES INVOLVED IN SUBSPECIES RECOGNITION AND SEXUAL ISOLATION IN THE HOUSE MOUSE: INSIGHTS FROM A TRANSCRIPTOMIC AND GENOMIC PERSPECTIVE

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The two subspecies of the house mouse Mus musculus musculus and M. m. domesticus have evolved in allopatry for half a million years before meeting secondarily in Europe 5000 years ago, forming a narrow hybrid zone with unfit hybrids. Previous studies have shown that these incipient species recognise each other through signals present in urine and mate assortatively in the contact zone. This suggests a reinforcement process in this zone where prezygotic isolation would evolve as a response to selection against hybridisation. Our study addresses the genetic basis of sexual isolation between these two subspecies by analyzing divergence of two large multigene families involved in chemosensory recognition. On the signaling side, we focused on the Major Urinary Proteins (MUP), a cluster of more than 21 duplicated genes known to act as pheromones in the house mouse. On the reception side, we focused on their potential receptors, the vomeronasal receptors (VR), a very large multigene family composed of more than 200 genes, expressed in the vomeronasal organ of the mouse and known to be involved in pheromone recognition. Since assortative mating is displayed in the hybrid zone, we predicted strong sign of divergence between the two subspecies at our candidate families provided that they are involved in sexual isolation. Moreover, if reinforcing selection is acting on the mate recognition system in the hybrid zone, we expected a stronger signature of selection in the genomic regions baring VR and MUP genes in individuals of the hybrid zone as compared to individuals residing in allopatric zones of the distribution range. By combining several high-throughput genomic methods such as RNA-seq and exome sequencing, we tested these predictions and explored divergence affecting the VR and MUP gene families at the sequence, expression and structural levels.

D20SY12PS1085

MHC VARIABILITY AND MATE CHOICE IN A RECOVERING WOLF POPULATION

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Isolated small populations might experience low adaptability and increased extinction risks due to reduced genetic variability, especially if at important functional genes such as the Major Histocompatibility Complex (MHC). We investigated for the first time the variability of three class-II genes (DRB1, DQA1, DQB1) in the Italian wolf population, which was long isolated and is now expanding after a recent bottleneck. Compared to other populations, we still found a remarkable MHC variability with 6 to 9 alleles per locus, showing signatures of historical selection (high dN/dS ratio). The Italian wolves are known to hybridize with domestic dogs, and dog-derived MHC alleles were detected in some genetically-admixed individuals. Background microsatellite and MHC loci did not show reduced variability due to the recent bottleneck. Thus, the population recovery seems not to be threatened by reduced MHC variation nor by deep domestic dog allele introgression. The observed variability could be also influenced by reproductive patterns, which can rely upon MHC similarity between mates. Based on pedigree data of wild-living packs, as reconstructed from extensive noninvasive genetic sampling, we tested the hypothesis of non-random mate choice in 26 breeding pairs. Unexpectedly, the results showed a prevalence of MHC-based assortative mating, with excess of peptide similarity between breeding pair members as compared to random expectations. Moreover, variation in both relatedness and heterozygosity showed significant positive correlations with fitness traits (total offspring, years as reproducers, mean offspring per year) deduced from pedigrees. These findings suggest possible advantages for breeders that are more related at the MHC, but not at the genetic background. This balance of general inbreeding avoidance, MHC-assortative mating and heterozygote advantage can reflect the social structure of the species, potentially maximizing the adaptation to the environmental pressures

D20SY12PS1138

LOW GENETIC DIVERSITY IN MHC LOCI IN THE CRITICALLY ENDANGERED IBERIAN LYNX

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A central premise in Conservation Genetics is that genetic erosion occurring in declining populations reduces adaptive potential and compromises viability. However, functional variation is seldom assessed in endangered populations. Here we use the Major Histocompatibility Complex class I and II-DRB loci (MHC) as functional model loci to study the impact of the decline on Iberian lynx genetic variation, a species for which previous genetic studies have shown a strong decline in the neutral variation and concomitant fitness reductions. We genotyped the most variable exon of MHC I and II-DRB genes in 300 individuals from the two extant wild and the captive populations using a 454 platform and a new pipeline analysis. We used transcriptomic and genomic data for primer design and expression analysis. Captive individuals, having a well-established pedigree, allowed us to infer composite haplotypes and to analyse segregation. We found a total of 13 and 6 alleles probably corresponding to 7 and 3 loci for MHCI and MHCII, respectively. One allele shows signals of pseudogenization and not all other alleles were confirmed to be expressed. In the case of MHCII, most individuals in the smallest and less diverse Doñana population show a three-allele profile, consistent with homozygosity at all three loci. Additionally global diversity levels for MCH_I and II are low compared to other felid species. Our results suggest that the immune response in remnant Iberian lynx populations may be compromised and that the severe reduction and fragmentation may have eroded not only the neutral, but also the functional genomic diversity, thus compromising the evolutionary adaptive potential of the species.

D20SY12PS1170

MHC-BASED QUALITATIVE AND QUANTITATIVE RESISTANCE TO AVIAN MALARIA IN A WILD BIRD POPULATION

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Major histocompatibility complex (Mhc) genes are believed to play a key role in the genetic basis of disease control. Although numerous studies have sought links between Mhc and disease prevalence, many have ignored the ecological and epidemiological aspects of the host-parasite interaction. Consequently, interpreting associations between prevalence and Mhc is difficult, while discriminating alleles for qualitative resistance, quantitative resistance and susceptibility remains challenging. Moreover, most studies to date have quantified associations between genotypes and disease status, overlooking the complex relationship between genotype and the properties of the Mhc molecule that interacts with parasites. Here, we solve these problems and demonstrate malaria parasite species-specific associations with functional properties of Mhc molecules in a wild bird population; effects that depend crucially on understanding the impacts and spatial epidemiology of these parasites. Furthermore, we demonstrate common functional properties of malaria-resistance alleles in passerines, suggesting this is a model system for parasite-Mhc associations in the wild.

D20SY12PS1309

THE INFLUENCE OF MHC ON FERTILITY SUCCESS IN HOUSE SPARROWS (PASSER DOMESTICUS)

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Over 30 years ago, the first study concerning MHC (major histocompatibility complex)-dependent mate choice was published in which female mice preferred MHC-dissimilar males. Since then, numerous studies about MHC preferences and mate choice in different species were issued. The MHC has a crucial role in the immune system and should therefore be important in mate choice decisions. There is evidence that breeding pairs who do not share the same MHC alleles are more 'compatible' in terms of fertility success and offspring survival. Most passer species are socially monogamous, but genetic polyandry is widespread. Females commonly engage in extra-pair matings and therefore it is likely that paternity of offspring is determined by postcopulatory sexual selection mechanisms. In this study we tested the postcopulatory fertility success of similar/dissimilar breeding pairs (respective to their MHC composition) in house sparrows. Additionally we analyzed if 'compatible' pairs also produce more chicks. With next generation sequencing technologies (454 sequencing), we sequenced the alleles of MHC class I, exon 3. We used an aviary population of wild house sparrows (61 males and 58 females) and kept them in 14 aviaries with 3-5 breeding pairs/aviary. The obvious benefit of an aviary population is that we could exclude several environmental factors influencing the results. We removed the first clutch of all breeding individuals (52 clutches with 4 eggs on average; 208 eggs total) to count the number of spermatozoa on the inner and outer perivitelline layer of the egg to study the postcopulatory fertility success. Additionally we allowed them to breed a second time to compare the postcopulatory success with the actual breeding success of these pairs (number of eggs, amount of hatching and fledging chicks). This comparison helped us to gain a deeper insight in the principles of MHC-dependent compatibility and fertility success.

D20SY12PS1373

VARIATION IN MHC II GENES IN THE CONTEXT OF GALAPAGOS MOCKINGBIRDS STUDY SYSTEM

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Galapagos mockingbirds comprise four species that radiated on the archipelago approximately 1.6-5.5 million years ago. Previous studies on mtDNA and microsatellite diversity as well as on co-evolution with their parasites showed geographically determined pattern of population structure, and differing levels of parasite load between some islands. Such well-studied system of Galapagos mockingbirds provides a robust background for answering questions related to MHC evolution in small populations of island colonists. Here we present preliminary results from an analysis of variation in the MHC class II genes, and hypotheses that we want to test when the genotyping is completed for a sample set of 250+ individuals. Using 454 method we sequenced exon 2B of the MHC class II in 33 individuals of Galapagos mockingbirds from 9 island populations. Substantial level of variation in this region was found, which is consistent with results published for other passerines. Number of variants per amplicon was on average 10.2 (S.D. = 3.3) with a coverage of 732 (S.D.= 297) reads per amplicon. As was expected, allelic variants were mostly shared between individuals from the same island or species, but a few exceptions occurred. Analyses of selection found high level of nonsynonymous mutations at multiple aminoacid positions. The questions to be tested in future work concern 1) the impact of drift and balancing selection on variation in the MHC gene alleles, 2) the relationship between MHC variation and ectoparasite load, 3) the usability of MHC in conservation genetics of the critically endangered Floreana mockingbird. Genetic compatibility of the two surviving populations will be analyzed. This will provide information useful to devise a plan of successful cross-breeding and reintroduction of the birds to mainland Floreana.

Symposium

13. Rapid Evolution and Population Genetics

21 August



Program

Wednesday 21 August

Session(s): 5, 6, 7, 8

Organisers: Lutz Becks and Teppo Hiltunen

Invited speakers: Hildegard Uecker and Richard Neher

Description:

While the synthesis of the so-called eco-evolutionary dynamics has seen large advances in the last decades, there is still further need to illuminate the relationship between ecology and evolution. In particular, we believe that the integration of rapid, contemporary evolution into studies of population genetics has largely been ignored.

D21SY13IT15:45R3

OBSERVING EVOLUTION IN ACTION: HIV AND ITS COEVOLUTION WITH THE HOST IMMUNE SYSTEM

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The immune system of a HIV infected individual is constantly fighting the virus. In absence of treatment, this battle is ultimately lost in the great majority of cases since the virus changes rapidly to evade the immune pressure. I will show how relevant evolutionary parameters such as typical selection coefficients and recombination rates can estimated from time series data of HIV. Standard population genetic methods are often inappropriate to analyze HIV data, since the viral population experiences strong positive selection at many sites. This implies that the relevant stochastic force in HIV evolution is not genetic drift but selection on linked sites or genetic draft and methods based on the neutral coalescent do not work. I will then discuss recent progress in developing a theory of rapidly adapting populations such as HIV. In models of rapid adaptation, genealogies are different from the standard Kingman coalescent but display frequent multi-merger event. Their statistic is described by the Bolthausen-Sznitman coalescent. These results allow us to calculate levels of neutral diversity, site frequency spectra, and the scale of linkage disequilibrium from parameters of the model.

D21SY13IT17:45R3

EVOLUTIONARY RESCUE IN STRUCTURED POPULATIONS

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As a consequence of environmental deterioration, a population might become maladapted and risk extinction unless it succeeds in adapting to the new conditions. How likely is it that a population escapes extinction through adaptive evolution? Modeling a population in a degrading structured habitat, we analyze the impact of several ecological factors on its survival probability and determine the relative contribution of standing genetic variation and new mutations to evolutionary rescue. We find that in the interplay of various, partially antagonistic effects, the probability of evolutionary rescue can show non-trivial and unexpected dependence on ecological characteristics. The rate of gene flow affects the fate of the population in several ways, resulting in a complex and non-monotonic relationship between migration rate and rescue probability. Counterintuitively, a harsher change or an instantaneous degradation of the total habitat can sometimes lead to a higher survival probability than a less severe or a slowly progressing change.

D21SY13RT10:30R8

WHAT LIMITS AN EVOLUTIONARY RESPONSE TO ENVIRONMENTAL CHANGE?

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Understanding why populations fail to adapt to environmental change is critical to explain the formation of species' range limits, and to predict the conditions under which populations will be able to evolve in response to climate change and habitat loss. Two hypotheses have been proposed to explain why adaptation fails in response to environmental change: (1) A lack of additive genetic variance in key traits or trait combinations inhibits a response to selection, and (2) Local adaptation in peripheral populations is swamped by gene flow from larger populations adapted to conditions at the centre of the range. Several theoretical models provide insights into the circumstances under which adaptation is expected to occur, but empirical tests of these predictions in wild populations are rare. We examined spatial changes in the mean and additive genetic variance of ecologically important traits of the rainforest specialist Drosophila birchii along repeated altitudinal gradients, to evaluate evidence for these competing hypotheses about limits to adaptation. We found little evidence for adaptive divergence in stress traits (cold tolerance, heat tolerance, desiccation resistance) or morphological traits (thorax length, wing size) at any transect, despite abundant genetic variance in most traits and populations. At two transects, male reproductive success was higher in high altitude populations than low altitude populations. Our results suggest that gene flow along altitudinal gradients is likely to be an important factor limiting adaptive divergence. However, we are also investigating the possibility that trade-offs between stress tolerance and reproductive success may constrain evolution at the range margins of this species.

D21SY13RT10:54R8

WHEN DOES GENETIC DRIFT PREVENT EXPANSION OF A SPECIES' RANGE?

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Genetic variance is crucial for adaptation-driven range expansion. Locally, genetic variance will be reduced by genetic drift. Assuming variance is free to evolve, when do low population numbers limit further expansion of a species' range? We employed the understanding of i) the deterministic dynamics of the joint change in population density and evolution of a quantitative trait and ii) the effect of genetic drift on a cline, to find a threshold where a finite population ceases to adapt to an environment changing in space. Clines in allele frequencies become steeper due to genetic drift and therefore local genetic variance decreases. This decrease in variance can lead to a sudden collapse of species' range, as variance (standing load) becomes too small relative to the steepness of the environmental gradient (dispersal load). Hence, as drift constraints the genetic variance in finite populations, even a small increase in the gradient of the environmental optimum can lead to a sudden inability of a species to continuously adapt to gradually changing environments, and drive a sharp range margin or range fragmentation.

D21SY13RT11:18R8

RAPID EVOLUTION IN EXPERIMENTALLY-HARVESTED FISH POPULATIONS

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Rapid contemporary evolution has become more common place in response to human-induced pressures such as introductions and overexploitation. One such increasingly significant pressure arises from size-selective harvesting in commercial fisheries which can induce rapid changes in biological traits. While experimental and wild harvested populations often show clear shifts in body size and maturation associated with fishing, the relative contributions of genetic and environmental factors to these shifts remain uncertain and have been much debated. Observations of so-called fisheries-induced evolution (FIE), to date are based solely on phenotypic measures, such as size data, and no genetic data are available. Here, we quantify genetic versus environmental change in response to size-selective harvesting in guppies (*Poecilia reticulata*) across three generations of selection for small and large body-sized individuals. We document for the first time significant changes at individual genetic loci, some of which have previously been associated with body size. In contrast, variation at neutral microsatellite markers was unaffected by selection, providing direct genetic evidence for rapid evolution induced by size-selective harvesting. These findings demonstrate FIE in an experimental system, with major implications for the sustainability of harvested populations, as well as impacts on size-structured communities and ecosystem processes. Such findings prompt scientists and managers to reconsider the capacity of harvested stocks to adapt to, and recover from, harvesting and predation.

D21SY13RT11:42R8

USING SEEDBANKS TO INVESTIGATE CONTEMPORARY EVOLUTION OF PLANT REPRODUCTIVE TRAITS FACING GLOBAL CHANGES

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Global changes raise numerous concerns about the fate of natural populations. Beyond ecological responses, global changes are also expected to impose new selective pressures on populations and hence represent an unprecedented opportunity to study evolutionary processes at contemporary scales. As a facet of global changes, pollinator decline has been reported at the global scale. Because plants usually depend on pollinators for their reproduction, plant reproductive traits (mating systems and floral traits) are expected to adapt to pollinator decline. Detecting contemporary evolution of natural populations is however a complicated task. For instance, advance in phenology has been reported for numerous plant species (from weeds to trees) in response to climate change, but it remains to be determined whether the mechanisms for such changes involve genetic evolution or reflect plasticity only. In our study we took advantage of the possibility to store viable seeds on a long period of time to analyze evolutionary changes from temporal samples. We benefit from seed conservation in optimal conditions by French Botanical Gardens to investigate rapid evolution of reproductive trait of the cornflower (Centaurea cyanus, Fam. Asteraceae), a self-incompatible crop weed. We study two generations of the same population (20 years apart) from a region where important reduction of both pollinator richness and density has been documented. Common garden experiments allow us to detect the effect of genetic evolution for major reproductive traits. Compared to the ancestral population, our results show that today's, plants flower earlier but also produce bigger flowers with longer receptivity, and exhibit bigger floral display. We used neutral markers to sort out the possibility that such differentiation in reproductive traits be due to drift only. We propose that such changes be in part adaptation to pollinator decline, through increased pollinator attraction.

D21SY13RT14:00R8

RAPID PHENOTYPIC AND GENETIC CHANGES IN EXPERIMENTALLY EXPLOITED FISH POPULATIONS

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Size-selective fishing has the potential to cause changes in life-history traits, which are often at least moderately heritable. Thus, size-selective fishing has the potential to cause genetic changes in exploited populations. We subjected populations of wild zebrafish (Danio rerio) to size-selective harvest over five generations by establishing three treatment groups: selection for large, random and small body size and focused on three questions that have not been thus far studied experimentally in fisheries context: 1) How do life-history traits, such as maturation, growth and reproductive investment, collectively respond to size-selective harvesting? 2) How the potential changes in life-history traits affect population growth rates? 3) Can size-selective harvesting induce rapid molecular-level responses in the experimentally exploited populations? Five generations of size-selective harvesting caused lower maximum length, earlier maturity and higher energy investment to reproduction among the small-size selected fish adapted to size-selective fishing environment and had higher population growth rate compared to other selection treatments. However, they had lower population growth rate in the absence of fishing. Thus, our results suggest that exploited populations

might not only be unprofitable for fisheries but also impair the recovery potential of exploited stocks. Finally, we show that size-selective harvesting induced rapid and replicable genetic changes in the experimentally exploited fish populations. Our study contributes to the understanding of the rate of phenotypic and evolutionary changes in size-selective fisheries. Further, our results suggest that fisheries-induced evolution is not necessarily a long-term process but it can be rapid and may become a short-term concern in fisheries management.

D21SY13RT14:24R8

QUICK ADAPTATION TO A NEW ENVIRONMENT ERASES SIGNATURE OF HISTORY IN NATURAL POPULATIONS

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In a world where human activity is changing the climate and habitats at a fast pace it is fundamental to understand how much and how quickly can species adapt. In the last few years we have witnessed the evolutionary response of various species to the effects of global warming. One such case is Drosophila subobscura, which presents clinal variation for body size and inversions, across three continents. Recent evidence shows that temporal changes are occurring in the clinal variation as a response to global warming, with northern populations becoming more similar to southern ones. Both local adaptation and gene flow may be involved, the latter possibly overcoming historical constraints. In this study we propose to measure the contributions of history and selection when populations initially differentiated in Nature are under a similar selective pressure, in order to test if the uniform selection erases prior genetic differences, even in the absence of gene flow. We used as scenario the adaptation to a new, common environment of three populations of Drosophila subobscura initially differentiated along the European latitudinal cline. Quick evolutionary response was observed in all foundations leading to full convergence. All foundations converged to the same adaptive peak, although at different rates and through different paths, suggesting an overall smooth fitness landscape. We concluded that although history had a strong effect during the initial generations, selection quickly overcame it, especially in fitness related traits. The fast loss of differentiation shows that, even in the absence of gene flow adaptation to a common environment can erase the variation observed in nature, a finding that raises concerns in Conservation terms.

D21SY13RT14:48R8

RAPID EVOLUTION OF METAL TOLERANCE REVEALED BY RESURRECTING DORMANT PROPAGULES

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We investigated the evolutionary response of an ecologically important freshwater crustacean, Daphnia, to a rapidly changing toxin environment. From the 1920s until the 1960s, the use of leaded gasoline caused the aquatic concentration of this toxin to increase at least 5-fold, presumably exerting rapid selective pressure on aquatic organisms to develop resistance. Similarly, we predicted that the banning of leaded gasoline, and the corresponding return to low lead levels, would result in the loss of resistance if this resistance carried a cost. These questions were addressed directly by using the resurrection ecology approach, whereby dormant propagules from a focal time period are hatched or germinated and compared to those from another time period. We hatched several Daphnia genotypes from each of two Swiss lakes, during times of higher (1960s / 1980s) and lower (2000s) lead stress, and compared their life histories under different laboratory levels of this stressor. The results were clear: modern Daphnia genotypes (hatched from a period of low lead pollution) had significantly reduced fitness, measured as the population growth rate (r), when exposed to lead, while those genotypes hatched from times of high lead pollution did not display this reduction. We conclude that Daphnia in these lakes were able to rapidly adapt to increasing lead concentrations, and just as rapidly lost this adaptation when the stressor was removed. These results are analyzed in context of the recent theoretical advances by Ellner et al. (2011), who developed a framework for the explicit comparison of ecological and evolutionary rates, and are used to demonstrate a previously unrecognized feature of their model. We also discuss avenues of further research into the genetic underpinnings of this adaptation, taking into account recent research on ABC transporters and technological advances in sequencing technology.

D21SY13RT15:12R8

HOST-PARASITE COEVOLUTION AT ECOLOGICAL TIME SCALES: BACTERIA-PHAGE DYNAMICS WITHIN A LONG-LIVED HOST

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Plants and animals host a diverse microbiota and it is increasingly clear that these dynamic microbial communities significantly affect host phenotype, including resistance to disease. Microbiota act as complex coevolving networks, with high levels of horizontal gene transfer, immigration of new species, selection by the host immune system, and multifaceted antagonistic interactions. A key challenge is to understand how microbial communities are influenced by interactions with their hosts and viral parasites (phages), and how these communities affect the fitness of their hosts. Given their sessile life histories, high risk of infection by microbes, and reliance on commensal microbial species, plant hosts represent an ideal system for testing both fundamental and applied questions in this emerging field. I first determined the relevant ecological structure of phage adaptation in natural microbial communities living within tree hosts and then measured the coevolutionary interaction between phages and bacterial communities within these tree hosts over time. I took an experimental time-shift approach and demonstrate that bacteria are most resistant to phages from the past and least resistant to those from the future. These results provide the first evidence that natural bacterial populations respond rapidly to local phage-mediated selection and that phages play a key role in shaping the microbiota of their eukaryotic hosts.

D21SY13RT16:33R3

RAPID EVOLUTION OF INSECTICIDE RESISTANCE GENES IN MOSQUITO POPULATIONS: A QUANTITATIVE APPROACH

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How adaptation appears and is later refined by natural selection has been the object of intense theoretical work. However, the testing of these theories is limited by our ability to estimate the strength of natural selection in nature. Selection by insecticide treatments of the Ester resistance gene in populations of the mosquito *Culex pipiens* in Montpellier area (southern France) is a contemporary example of rapid evolution of an adaptation to environmental changes in natural populations. We observed in twenty years, the emergence and replacement of three resistance alleles at the Ester locus due to the selection pressure of insecticide uses. With continuous sampling for >40 years on a 50 km transect corresponding to a gradient of insecticides treatment, we were able to follow the dynamics of these different alleles in natural populations. Using a population genetics model taking into account environmental variables, we were able to estimate the intensity of the selection pressure, as well as many parameters (genetic and environmental) affecting the dynamics of these alleles. In addition, we were able to analyze the evolution of allele frequencies under various environmental conditions. For example the prohibition by the European instances of OP's insecticides in 2007 was a full-scale test of the quality of our predictions. I will present this work, which links ecology and evolution, through the integration of environmental data into a genetic model, to analyze the dynamics of adaptive alleles in natural populations. We quantified the selection and environmental parameters conditioning resistance genes dynamic and showed that the relationship between environmental factors and allele dynamics in natural populations is not straightforward.

D21SY13RT16:57R3

DNA SEQUENCE RESPONSE TO EXPERIMENTAL EVOLUTION AT DIFFERENT SEXUAL SELECTION CONDITIONS

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We have been maintaining *Drosophila pseudoobscura* populations in two regimes of high and low sexual selection for 150 generations. Several phenotypes have responded to selection during this limited time, for example courtship behaviour, morphology, fecundity and chemical communication. We conducted RAD sequencing of 4 biological replicates of each condition and obtained >7000 regions containing SNP variation. We characterise the genes and genomic regions associated with consistent differences in SNPs between the selection regimes. We also comment on the repeatability of the response, with comparisons within each biological replicate.

D21SY13RT18:33R3

ENVIRONMENTAL VARIATION AND THE EVOLUTION OF VIRULENCE

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Environmental opportunist pathogens are common in nature. Two well known examples are Vibrio cholera in humans and Flavobacteria in fish. Yet studies on epidemiology and evolution of pathogenicity are centred on obligate pathogens. Environmental opportunist pathogens do not require host-to-host contacts for transmission, and spend most of their time in the outside-host environment where their abundance depends on biotic and abiotic conditions. Such environmental bacteria can spontaneously gain and loose virulence factors that are likely to be associated with increased growth potential and energetic costs. Non-obligate pathogens have not tightly coevolved to evade host immune system, and pathogen transmission is likely to be strongly dose-dependent. This leads to a paradox: in the absence of hosts pathogenicity is selected against due to reduced competitive ability and this in turn prevents transmission to hosts. How can environmental opportunistic pathogens exist? We propose that environmental variation is a plausible mechanism explaining transition to more virulent forms. We present a model which combines a SIR system to environmental virulent and non-virulent bacterial strains. We assume the pathogenic strain is a fast growing whereas the competing strain is a better competitor. This system is associated with bi-stability between two regimes: virulent strain with infections and non-virulent strain with no infections. Starting from a lower density the virulent strain is unable to grow or cause infections in a stable environment. If both bacterial strains are subjected to environmental variation, the virulent strain can overcome between-strain competition and increase in density close to infective dose and cause sporadic outbreaks or persistent infections, and thus gain significant fitness increases. Infective cycles may in turn promote further evolution from environmental opportunism to obligate pathogenicity, especially in the case of persistent infections.

D21SY13RT18:57R3

EVOLUTION OF EVOLVABILITY UNDER FLUCTUATING SELECTION

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Empirical evidence suggests that fluctuating selection is a major evolutionary mechanism. The most straightforward consequence of rapid changes of the fitness function is the induced response of the mean phenotype in the population. Yet, repeated back-and-forth evolutionary trajectories are also suspected to affect the genetic architecture underlying the phenotypic characters subject to continuous adaptation. In order to better understand the long-term consequences of fluctuating selection, we modeled the response of complex, multilocus genetic architectures to various natural selection regimes -- stabilizing, directional, and fluctuating. This model accounts for gene-gene interactions (through multilinear epistasis), and thus allows to investigate the dynamics of evolutionary potential at two distinct levels: (i) the standing genetic variation, i.e. the capacity for the population to respond immediately to directional selection, and (ii) the level of canalization (measured as the average effect of new mutations), which reflects the capacity for the population to replenish genetic variation. Both analytical results and individual-based simulations show that fast fluctuations (white noise change in the phenotypic optimum every generation) are essentially similar to stabilizing selection, promoting a degree of genetic canalization and low evolvability. In contrast, when large fluctuations of the phenotypic optimum (beyond the phenotypic range of the population) occur every 10 to 100 generations, equilibrium mutational effects and genetic variance are higher and the population is more evolvable. However, there was no evidence that decanalization and increased evolvability were adaptive, and fluctuating selection remains intrinsically more constraining than genetic drift.

POSTERS

D21SY13PS0001

STRESS-INDUCED MUTATION IN THE PRESENCE OF RECOMBINATION

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Mutation is a key feature of every biological system and the evolution of the mutation rate is the subject of many theoretical and experimental studies. A major factor complicating our understanding of the evolution of the mutation rate is recombination: it separates mutator alleles from the beneficial mutations they generate and accelerates the fixation of beneficial mutations, but it also purges deleterious mutations and generates beneficial allele combinations lost by mutation and drift. Here we present an evolutionary model in which stress induces increased mutation rates. We show that this induction can evolve in the presence of rare recombination, such as experienced by bacteria and yeast, and explore the effect of stress-induced mutation on the evolution of recombination due to a process known as *Muller's ratchet*.

D21SY13PS0044

COULD RAPID ADAPTATION BE CONTRIBUTING TO THE SUCCESS OF INVASIVE SENECIO MADAGASCARIENSIS IN AUSTRALIA? COMBINING OUTLIER AND SPATIAL ANALYSIS METHODS TO DETECT PUTATIVE SELECTION

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Identifying contemporary evolution is a major goal in invasion biology. A first step in this endeavour is to identify loci which may have been subject to recent selection and show associations with particular environmental conditions. Here we used 164 amplified fragment length polymorphism (AFLP) loci to carry out genome scans on a total of 316 individual Senecio madagascariensis plants collected from across the known invaded range in Australia. Half of these (n = 158) were collected from eight populations and the remainder collected as individual plants >2 km apart. The population and individual based samples were analysed separately using two complementary approaches for identifying putative loci under selection. Populations were analysed using outlier approaches in two programs MCHEZA and BAYESCAN; individuals were analysed using a spatial analysis method in SAM to look for associations between particular loci and environmental variables. Two loci were identified in both datasets as potentially under selection and were associated with monthly environmental variables describing light and water availability between May and August which corresponds with flowering time. Overall 6% of loci were identified as candidates for selection in one or more of the analysis methods, all three software programs only agreed on a single locus (0.6% of all loci) and this was associated with eight environmental parameters describing rainfall. Results are discussed with reference to selection and alternative explanations for the observed patterns. The two identified loci present promising targets for future investigations into contemporary selection in invasive S. madagascariensis in Australia.

POPULATION GENETIC STRUCTURE OF WATER FLEA (DAPHNIA PULEX SPP.) FROM GRADIENTS OF LAKES AND PONDS IN ILULISSAT REGION OF GREENLAND

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Daphnia has quickly emerged into model organism in ecology and evolution research. As a result it has been subjected to extensive studies in eco-toxicology, ecological genomics and population genetics studies. However, most of the population genetics studies of Daphnia have concentrated in temperate population and a few from arctic Canada. Given to its sensitivity to climate change, the expansion of such study into the polar region is of paramount importance. What is more, the study of population structure and diversity of organisms from the polar region can provide a unique opportunity to make inferences about postglacial colonization in areas that have recently been de-glaciated due to climate change and contribute to our understanding of colonization dynamics and founder events. Here, we assessed the spatial population genetic structure of Daphnia pulex spp. complex from a gradient of lakes and ponds in Greenland, which are recently created due to melting of the Greenland Ice Sheet. 61 population were sampled from 21 lakes and 40 ponds that vary in area, size and depth and genotyped at 9 microsatellite loci. Based on 9 microsatellite loci, we scored 1-9 MLGs/population which vary in their distribution, clonal diversity and mean clonal richness in each habitat studied. Clonal diversity ranges 1.22 to 3.65 and 1.22 to 3.72 for Lakes and ponds respectively. 20-90% expected heterozygosity was observed for both lakes and ponds. Furthermore, these ponds/lakes harbor two/more of the polyploidy members of Daphnia pulex spp. (i.e. Daphnia pulicaria, and Daphnia middendorffiana). However, Daphnia pulicaria seems to dominate [i.e. having high frequency of occurrence] compared to Daphnia middendorffiana. In general, our finding support hypothesis that polyploidy is dominant in marginalized and higher latitude. But this needs further confirmation by increasing the number individuals genotyped in a population and genetic marker used.

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LOTKA AND VOLTERRA KILL THE RED QUEEN

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Host-parasite coevolution is generally believed to follow so-called Red Queen dynamics consisting of ongoing oscillations in the frequencies of interacting host and parasite alleles. This view is based on current empirical and theoretical work, which specifically focuses on the evolutionary dynamics of these interactions (i.e., the change in host and parasite allele frequencies). Here, we demonstrate that consideration of a central ecological component, population size fluctuations, dramatically alters these dynamics. In particular, reciprocal selection among antagonists causes inter-dependent demographic variations, as described by the Lotka-Volterra relationship. These population size fluctuations are thus an inherent characteristic of host-parasite coevolution. As they associate with repeated bottlenecks, they increase the likelihood of genetic drift. This in turn favours fast fixation of one of the interacting alleles, often including the allele that is originally rare. As a consequence, any ongoing Red Queen dynamics is rapidly terminated. Taken together, our results suggest that host-parasite coevolution is mainly driven by selective sweeps rather than continuous negative frequency-dependent selection. Long-term coevolution consistent with the Red Queen hypothesis is only possible if new alleles are repeatedly introduced *de novo* into the interacting host and parasite populations.

COMPARING EVOLVABILITIES: COMMON ERRORS SURROUNDING THE CALCULATION AND USE OF COEFFICIENTS OF ADDITIVE GENETIC VARIATION

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Evolvability, the ability of populations to respond to natural or sexual selection, is contingent on the level of additive genetic variation underlying trait expression. Two valuable evolvability measures are the coefficient of additive genetic variation (CVA) and its square (IA), which standardize the additive genetic variance by the mean of the trait and therefore allow comparisons among traits and taxa. CVA has been used widely to compare patterns of genetic variation. However, the utility of CVAs relies critically on the correct calculation of this parameter. We reviewed a sample of quantitative genetic studies, focusing on full sib-half sib breeding designs, and found that 45% of studies use incorrect methods for calculating CVA and that practices that render these coefficients meaningless are frequent. This is likely to compromise studies that use such statistics for comparative purposes. Our results are suggestive of a broader problem because miscalculation of the additive genetic variance from a sire model is prevalent among the studies sampled, implying that other important quantitative genetic parameters might also often be estimated incorrectly. Here we outline some issues affecting the use of CVA and IA, including scale effects, data transformation, and the comparison of traits with different dimensions. We also compile general guidelines for calculating, reporting and interpreting these evolvability measures in future studies.

COMPARISON OF HAPLOTYPE METHODS TO DETECT SELECTION

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Motivation: The main topic of research in human genetics is the identification of genes and mutations that contribute to a genetic disease. One of the factors that can influence the genetic diversity in a population is natural selection. In this work, we will compare the existing long-range haplotype methods to detect selection. Our primary objective is to obtain a clear view about their power and validity under complex demographic scenarios. Methods & Results: Literature review: A systematic review that was conducted revealed five haplotype methods with available software to identify loci that have undergone selection. The five methods are the following: LRH (Sabeti, 2002; Nature 419:832-837), iHS (Voight, 2006; PLos Biol 4:72), xp-EHH (Sabeti, 2007; Nature 449:913-8), EHHST (Zhong, 2010; Hum Genet 18:1148-59) and xp-EHHST (Zhong, 2011; Stat & Its Interface 4:51-63). All of them used simulations to test their performance with the ms (Hudson, 2002; Bioinf 18:337-8) and SelSim programs (Spencer, 2004; Bioinf 20:3673-5). Sensitivity Analysis: Ms program can consider different demographic models, without selection and SelSim provides simulations under natural selection with a simple population structure. To determine the best method, we will generate simulated data using SimuPOP. SimuPOP is a forward-in-time simulation program that can construct models with selection under complex evolutionary scenarios. We will begin with an island model, a stepping stone model incorporating an environmental gradient and more complex scenarios including a hierarchically structured population. Conclusions: We will thoroughly investigate their behavior under complex scenarios. Our study sets the basis to identify the advantages and disadvantages of each method under each modeling assumption. Here, we restrict ourselves to the comparison of the methods but an extension to a model to detect selection to N populations could be developed at a later stage.

INCIPIENT ALLOCHRONIC SPECIATION IN THE PINE PROCESSIONARY MOTH : MOLECULAR ANALYSIS OF A DIVERGENT POPULATION

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A case of allochronic differentiation, where genetic isolation is due to a shift in the reproductive period, has been shown recently in the pine processionary moth (PPM; Thaumetopoea pityocampa) in Portugal. The PPM is a Mediterranean Lepidoptera which reproduces in summer and whose larvae develop during winter. A population with a "shifted" phenology, hereafter named the "summer population" (SP) referring to its period of larval development, has been discovered in a coastal maritime pine forest in sympatry with a population showing a "classic" phenology, hereafter the "winter population" (WP). The phenological shift is such that sexual reproduction seems to not overlap in the field between the SP and the WP. A previous study based on a limited number of markers proposed a local and recent origin of the SP from the sympatric WP. We aimed at defining the demogenetic history of these two populations thanks to genotypic data from 19 microsatellites markers and mitochondrial DNA sequences (COI gene). Genetic and phylogeographic analyses confirmed the local origin of the SP and showed a strong differentiation of this population from those with a "classic" phenology. These results also identified both genetically SP individuals who came back to a WP life cycle and possible hybrid individuals, assuming the possibility of gene flow between the two divergent populations. We suggested several demographic scenarios describing the evolutionary history of the SP and the WP. These scenarios have been tested using a Bayesian inference method, namely Approximate Bayesian Computation (ABC). The models tested did not allow to perfectly describe the observed data and should still be improved. New, complementary genetic markers (SNPs) will be developed to attain this goal.

A TWO-FOLD ADVANTAGE OF SEX

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The adaptation of large asexual populations is hampered by the competition between independently arising beneficial mutations in different individuals, which is known as clonal interference. In classic work, Fisher and Muller proposed that recombination provides an evolutionary advantage in large populations by alleviating this competition. Based on recent progress in quantifying the speed of adaptation in asexual populations undergoing clonal interference, we present a detailed analysis of the Fisher-Muller mechanism for a model genome consisting of two loci with an infinite number of beneficial alleles each and multiplicative (non-epistatic) fitness effects. We solve the deterministic, infinite population dynamics exactly and show that, for a particular, natural mutation scheme, the speed of adaptation in sexuals is twice as large as in asexuals. This result is argued to hold for any nonzero value of the rate of recombination. Guided by the infinite population result and by previous work on asexual adaptation, we postulate an expression for the speed of adaptation in finite sexual populations that agrees with numerical simulations over a wide range of population sizes and recombination rates. The ratio of the sexual to asexual adaptation speed is a function of population size that increases in the clonal interference regime and approaches 2 for extremely large populations. The simulations also show that recombination leads to a strong equalization of the number of fixed mutations in the two loci. The generalization of the model to an arbitrary number \$L\$ of loci is briefly discussed. For a particular communal recombination scheme, the ratio of the sexual to asexual adaptation speed is approximately equal to \$L\$ in large populations.

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ADVANTAGES OF SEX BEYOND RECOMBINATION IN A FUNGAL MODEL

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Why sexual reproduction is so prevalent in nature remains a major question in evolutionary biology. Most of the proposed advantages of sex rely on the benefits obtained from recombination. However, it is still unclear whether the conditions under which these recombinatorial benefits would be sufficient to maintain sex in the short term are met in nature. Our study addresses a largely overlooked hypothesis, proposing that sex could be maintained in the short term by advantages due to functions associated with sex, but not related to recombination. These advantages would be so essential that sex could not be lost in the short term. Here, we used the fungus Aspergillus nidulans to experimentally test predictions of this hypothesis. Specifically, we were interested in (1) the short-term deleterious effects of recombination (2) possible non-recombinatorial advantages of sex particularly through the elimination of mutations, and (3) the outcrossing rate under choice conditions in a haploid fungus able to reproduce by both: outcrossing and haploid-selfing. Our results were consistent with the hypothesis: we found that 1) recombination can be strongly deleterious in the short term; 2) sexual reproduction between individuals derived from the same clonal lineage provided non-recombinatorial advantages likely through a selection arena mechanism.

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HUMAN-ASSISTED ADMIXTURE AND INVASION – A CASE STUDY WITH ZANDER (SANDER LUCIOPERCA)

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Non-native species receive attention because they are perceived as a threat to native biodiversity. Besides ecological factors, evolutionary processes may determine the invasion potential of a species. Genetic admixture has the potential to induce rapid evolutionary change and can occur when secondary contact between differentiated populations is established. Here, we analyse population genetic signatures of admixture of zander (Sander lucioperca) and test the hypothesis that the invasion potential of non-native populations was enhanced by admixture. We use zander as a model system whose range expansion was facilitated by human transfer in the last century in Germany. The rivers Elbe and Danube harboured the most western populations representing source populations for nonnative areas west of the River Elbe and north of the Danube. Stocking and the connection of drainages through canals greatly facilitated dispersal and genetic admixture. We collected more than 1.300 specimens from the ancestral ranges as well as the newly colonized area in Germany and genotyped these using microsatellites and mitochondrial sequences. We studied the genetic population structure for signatures of admixture. Human-mediated secondary contact led to increased genetic diversity of zander via hybridisation among populations. We found populations of newly colonised areas to be more admixed than those of ancestral areas who maintained a high proportion of their ancestral genotypes. Admixture patterns were asymmetric and occurred mainly from ancestral populations to the newly colonized ranges but less so in the opposite direction. We suppose that the adaptive potential of the invading populations was promoted by genetic admixture. By contrast, local adaptation in native areas may have provided a buffer against invasion by novel genotypes. These explanations would be in line with evidence that hybridization can drive evolutionary change under conditions when new niches can be exploited.

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FIRST INSIGHTS INTO THE GENETIC DIVERSITY OF THE PINEWOOD NEMATODE BURSAPHELENCHUS XYLOPHILUS IN ITS NATIVE AREA AND AROUND THE WORLD

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Biological invasions provide a great opportunity for research in evolutionary biology and specifically in short-term evolutionary process. For example, understanding the factors of success or failure of introductions and establishments in remote areas can improve our knowledge on both adaptation dynamics and colonisation processes in the context of invasions. Identifying the invasion routes and determining the origin of new outbreaks are of crucial importance and are a pre-requisite to address these questions. This may also permit to improve or establish regulatory measures and to potentially limit the damage. The pinewood nematode (PWN), Bursaphelenchus xylophilus is the causal agent of the pine wilt disease and is currently considered as one of the most important pests and pathogens in forest ecosystems. Native to North America, it has been introduced and it has spread in pine forests in Asia and Europe where it has now considerable economic and environmental impacts. In order to decipher the invasion routes and to improve our knowledge about this specific case of invasion using population genetic approaches, we have developed a set of PWN-specific microsatellite markers, usable in routine conditions at the individual level. Preliminary results on a set of samples from the native (North America) and invasive (Europe and Asia) areas indicate: (i) a fine spatial genetic structure at the scale of the pine tree and probably at larger scales in the native area and (ii) a very low level of polymorphism in PWN populations from invasive areas. The genotyping of samples from more representative areas in Europe, Asia and North America is currently underway in the laboratory in order to confirm/refine these conclusions. Assessing the genetic diversity of populations constitutes the cornerstone to determine the source of the European invasive PWN populations and whether they are the result of a single or several independent events of introduction.

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CLINAL ANALYSIS OF A TEMPORARY CONTACT ZONE BETWEEN TWO INVADING OUTBREAKS TO ESTIMATE DISPERSAL OF A MAJOR PEST OF MAIZE

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Dispersal is a key parameter involved in invasion, persistence and evolution of species. Despite the interest of obtaining dispersal estimates, measuring dispersal remains a real challenge due to its complexity and associated technical constraints. Some biological invasions may, however, be useful to estimate the dispersal. The western corn rootworm (WCR), Diabrotica virgifera virgifera, is one of the most destructive pests of corn and is invading Europe. The two main European invasive outbreaks of WCR are located in North-Western Italy and Central Europe and they originated from independent introductions from North America. Recently it was demonstrated that a secondary contact probably occurred between these two expanding outbreaks in 2008, in Northern Italy and led to the formation of an admixture zone. Here, we exploit this ongoing process to infer the dispersal of the WCR by analysing temporary frequency clines at 13 microsatellite markers. A measure of the clines slope and the linkage disequilibrium between microsatellites at the centre of the contact zone provided two estimates of WCR dispersal. Simulations with various non Gaussian dispersal kernels and population density heterogeneity in space showed that these estimations are robust to several deviations from the diffusion approximation. The order of magnitude of this estimation is then compared to others estimates of WCR dispersal and is discussed in the context of biological invasions. We conclude that secondary contacts between colonizing populations, which are common in invasive species, are very useful and could be used to infer dispersal parameters in many species.

EVOLUTION OF ENVIRONMENTAL OPPORTUNIST PATHOGENS

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Most theories of the evolution of virulence concentrate on obligatory host-pathogen relationships. Yet, many pathogens are environmental opportunists that can also replicate and interact in the outside-host environment. As the survival of these pathogens can be host-independent, also the transmissionvirulence trade-off assumed in obligatory pathogens can be relaxed. This might promote evolution of virulence. In environmental opportunist pathogens, selection in the outside-host environment can influence the evolution of virulence and disease dynamics. There is evidence of a trade-off between capability to invade and live in the host, and the efficiency to use outside-host resources. Therefore environmental opportunist pathogens might be relatively weak outside-host competitors. How are new environmental opportunist pathogens then able to invade? We introduce a novel model that combines density-dependent growth and Lotka-Volterra competition between pathogen and non-pathogenic bacteria in the outside-host environment to SI host-pathogen dynamics. We studied evolution of pathogenicity as the ability for new environmental opportunist pathogens to invade. Parameterization was based on columnaris disease that is a worldwide nuisance of fresh water fishes in fish farms. New environmental opportunist pathogen is able to invade when host growth and outside-host growth of the pathogen are high enough to compensate lower outside-host competition ability. Also, increase in virulence promotes invasion. Selection could thus favor higher virulence in environmental opportunist diseases as compared to obligatory diseases. Strong outside-host competition on the other hand can drive new environmental opportunist pathogen to extinction. Therefore, situations where ecological constrains, such as competition, are relaxed promote environmental opportunism. Thus multiple outside-host ecological constrains, such as competition, can efficiently limit the emergence of new diseases.

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EVOLUTION OF INBREEDING AND OUTBREEDING DEPRESSION REVEALED IN 100 GENERATIONS OF LABORATORY ADAPTATION IN C. ELEGANS

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To understand the genetics underlying the evolution of inbreeding depression we performed experimental evolution in *Caenorhabditis elegans* for 100 generations under two different mating systems, where different inbreeding rates were imposed. Ancestral and evolved populations were tested upon enforced inbreeding for survival and also for the expected neutral diversity at 337 single-nucleotide polymorphism (SNPs) after inbreeding. Dioecious lineages inbred by sib mating underwent extinction at higher rates than androdioecious lineages inbred by selfing. In parallel, measurements of egg-to-adult viability over time showed that this fitness trait showed similar initial inbreeding depression at both mating systems, but different starting outbreeding depression. There was also evidence of selection for the maintenance of population structure during androdioecious evolution In contrast, and despite the complex dynamics, dioecious populations generally showed selection for population homogeneity. Our findings suggest that high inbreeding rates influence adaptation by efficient purging of deleterious recessive alleles which in turn create the opportunity for balancing selection. On the other hand, low inbreeding rates influence adaptation by the maintenance of deleterious recessive alleles and, possibly, by the maintenance of the locally-adapted sets of loci known to be common among wild *C. elegans* isolates.

NON DIRECTIONAL EPISTASIS INCREASES SELECTION LIMITS THROUGH ALLELIC REVERSALS

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Quantitave genetics commonly assumes that loci contribute independently to the phenotype. However, genetic interactions are pervasive in nature and the relevance of these interactions for the evolutionary process has been debated since the beginnings of the modern synthesis. We use a quantitative genetics framework with a pairwise model of globally non-directional epistasis to explore the relevance of these interactions under directional selection. We pay special attention to the dynamics of additive variance in the deterministic limit, and to the consequences of epistasis for the ultimate fate of individual alleles. We find that epistasis prolongs the response and increases selection limits only if it induces allelic reversals, i.e., if there is sign epistasis. Furthermore, we show how different allelic distributions in the source population and the details of their genetic architecture translate into different selection limits. Our results also suggest that architectures that evolved under stabilizing selection may display features that increase their potential for evolution when the trait becomes the target of directional selection. We then use a branching process formalism to show how the probability of fixation of a rare allele is affected by epistatic interactions with a series of deterministic sweeps. In particular, we show that the selection limit is also increased in this limit by an epistatic architecture, and that this effect is mediated by an increase in the number of alleles that ultimately display a positive probability of fixation.

ARTIFICIAL SELECTION FOR HOST-PLANT USE OF A SEED-PREDATOR: FITNESS CONSEQUENCES, INBREEDING DEPRESSION, AND GENETIC VARIATION

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The use of alternative, suboptimal food-plant species affect herbivore's fitness and life-history, and thus has both ecological and evolutionary consequences. Our study species, the seed predator Lygaeus *equestris* uses *Vincetoxicum hirundinaria* as its primary food plant. However, extensive spatio-temporal variation in seed production of *V. hirundinaria* occasionally enforces *L. equestris* to feed on alternative plant species. We conducted a long-term selection experiment to test if this seed predator is able to adapt to a suboptimal food-plant species in more than 20 generations. We measured fitness and adaptation in terms of increase in reproductive output. We replicated the populations within the selection lines to separate the effects of selection from random drift and conducted intra- and interpopulation crosses to detect inbreeding and population differentiation. Furthermore, we analysed how population genetic structure changed during the selection experiment. We found that although the fitness of *L. equestris* that had fed on the alternative food plant was almost seven times lower than when feeding on *V. hirundinaria*, it increased significantly during the experiment indicating selection for higher fitness on the alternative food plant. Besides selection, random drift affected adaptation to the alternative food plant as indicated by differences in fitness among the replicate populations. Interpopulation crosses within the selection lines resulted to higher fitness than intra-population crosses indicating inbreeding depression. To further underline the negative effects of random drift and inbreeding on adaptive potential of populations, we found that the level of genetic variation was lower in replicate populations feeding on the alternative food plant. Our study is novel in that it combines the analysis of population genetic structure to a more traditional selection experiment to examine hostplant specialization of herbivorous insects.

GENETIC DIFFERENTIATION OF SALMONIDAE POPULATIONS IN SOUTHERN BALTIC SEA IDENTIFIED FROM A SNP GENOTYPING

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Sea trout Salmo trutta and salmon Salmo salar belonging to Salmonidae family, have a wide distribution, including the rivers flowing into the Baltic Sea. Sea trout migrate from natal rivers to the sea where they forage until reaching sexual maturity, subsequently returning to their native rivers to spawn. The accessibility of natural spawning grounds has become very limited and most populations have been supported by stocking, resulting in a disrupted biodiversity. Genotyping of two Polish populations of sea trout from Vistula and Pomeranian river Słupia has been performed using an Atlantic salmon derived SNP-array. The average Fst over 39 polymorphic loci was 0.13. Generally, Vistula population was more genetically homogeneous, while the population of Pomeranian river was more diverse. Sixty SNPs were selected for further genetic studies of the Southern Baltic populations. Nine sea trout populations (from Poland, Lithuania, Bornholm, Russia and Estonia) were genotyped with the iPLEX Gold technology (Sequenom). Results indicated that the degree of genetic differentiation among populations, measured for 23 loci was moderate (average Fst=0.09). The high differentiation was observed between Polish river Słupia and Russian river (Fst=0.16). Low diversity was detected between Lithuanian Neman river and Polish Vistula river populations. Genetic structure analysis indicates that individuals from 9 populations were clustered into four groups. Atlantic salmon populations have a hierarchical structure of population differentiation and Baltic salmon forms one effectively isolated evolutionary unit of Atlantic salmon. Five populations of salmon (from Polish river Słupia and hatcheries, from Sweden and from Lithuania) were genotyped using an Atlantic salmon derived SNP-array (Illumina). Preliminary results indicated a close relationship between Polish populations from hatcheries relative to wild one and the distinctness of Swedish population.

POPULATION GENETICS OF TRANSCRIPTION BINDING SITES IN REGULATORY SEQUENCES

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Variation in protein coding regions cannot explain the extensive phenotypic diversity between and among populations. It is believed that variation in transcriptome levels which are regulated by noncoding DNA are responsible for natural variation. However, lack of a regulatory code prevents us from understanding the evolution of regulatory sequences. We would like to know what different patterns of regulation evolve in nature. For example, we would like to understand why we observe fuzzy promoters (i.e. many but not so strong protein bindings), or what turn-over rates are expected between functional regulatory sequences. We need to understand microscopic molecular mechanisms to entangle macroscopic evolutionary behaviors. In this study, we integrate a biophysical model of transcription into population genetics framework in order to understand the evolution of regulatory sequences. The biophysical model takes into account energy interactions among protein, RNApolymerase and DNA. It approximates the gene expression level with a thermodynamic approach. We consider finite haploid populations evolving under mutation and selection. We analyze both stationary and dynamical properties of evolved regulatory sequences. We show what microscopic (e.g. protein binding size) and macroscopic (e.g. population size) parameters affect the results. We discuss applications of our model to existing biological data.

13. Rapid Evolution and Population Genetics

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CHARACTERISTIC OF ATLANTIC COD (GADUS MORHUA, L.) POPULATIONS IN THE BALTIC SEA ACCORDING TO ITS ADAPTATIONS TO LOCAL ENVIRONMENTAL CHANGES

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Atlantic cod (*G. morhua*, L.) presently is one of the most valuable commercial species in the Baltic Sea. Cod's distribution in the Baltic Sea may depends on salinity and availability of spawning and feeding areas. Five samples of Baltic cod (n=150) were used for analyzing SNPs and genes expression. Data obtained for genes related to environmental conditions, e.g. salinity and temperature (alphaenolase (Gm1156), heat-shock protein 90 (Hsp90), aromatase (Aro9), and neuropeptide Y (NPY)) show different distribution of alleles and suggest presence of minimum 2 populations of cod: eastern and western. The transitions area between those populations are correlated to decrease of salinity. The eastern stock is adapted to local environment, characterized by very low salinity (under 7 PSU). In fish from western Baltic Sea we observe higher genetic heterogeneity than in the eastern population, which has a very low admixture of alleles other than those correlated with low salinity. Western population is probably under strong influence of the North Sea cod's stock. Further research should indicate if hypothesis assuming link between this differentiation and influence of local adaptations of cod is valid.

VARIATION IN GENE EXPRESSION DURING EMBRYOGENESIS IN DROSOPHILA STRAINS AND SPECIES

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In nature, we observe phenotypic variation within species as well as between species. The ratio of between-species to within-species variation can tell us about the type of evolutionary forces acting on the underlying genes. To discover how different genes involved in embryogenesis are evolving, we measured the variation of gene expression in early embryos of D. melanogaster inbred lines from North Carolina (DGRP). We focus on gene expression because we can directly analyse the effects of genetic variations in the regulatory elements. In this study, we analyse the amount of variation seen in inbred lines, and compare it to five of the sequenced Drosophila species. In addition, we estimate the importance of local adaptation by comparing D. melanogaster strains from North America with D. melanogaster strains from Africa. Our results show that there is substantial variation in gene expression during Drosophila embryogenesis, which is important for divergence between both populations and species.

SAPTIO-TEMPORAL GENETIC DIVERSITY OF BLOOMS OF ALEXANDRIUM MINUTUM

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Over the last decades, toxic blooms of the dinoflagellate Alexandrium minutum were recurrently observed along the Brittany coast (France). Bloom-forming phytoplanktonic species have been intensively studied regarding their ecology and the environmental conditions enhancing blooms formation. However, little is known about genetic diversity within these blooms. Phytoplankton is generally considered to have high dispersal ability and low genetic structure. The reproduction of dinoflagellates is not known in nature but in the lab both clonality and sexual reproduction are possible. It is generally assumed that bloom formation consists of only a few clones that multiply asexually, but recent population genetic studies demonstrated that it is probably not the case since high genetic diversity was found within natural populations (Richlen et al. 2012; Lebret et al. 2012). The aim of our study was to investigate spatio-temporal genetic diversity during blooms formation of Alexandrium minutum in two estuaries (Penzé and Rance). For this, we used 7 highly polymorphic microsatellites markers (developed by Nagai et al. 2006) from 283 monoclonal strains. Both sites were sampled at different dates during the bloom duration (days to weeks intervals during a month), in the two sites and two successive years. The results indicated that blooms of A. minutum showed a high genetic diversity suggesting that reproduction of this species is mainly sexual. Moreover, our results show that temporal variations within a bloom were higher than annual or spatial variations, this temporal genetic variation suggest that complex demographic processes are occurring within a bloom (i.e. population extinction, colonization either from sediments or outside the estuaries). We discuss how parasites might be responsible of these temporal dynamics. Keywords: Genetic diversity; genetic differentiation; microsatellites; HAB; Alexandrium minutum.

THE SIGNATURE OF PAST POPULATION ISOLATION ON GENE GENEALOGIES AND DNA SEQUENCE DATA

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Newly available genomic data are continuously challenging scientists and raising new questions, which require a deep understanding of the dynamics of DNA polymorphism in populations. The signature of past population expansion or bottleneck on gene genealogies and DNA sequence data have been well studied. However, other demographic events such as past isolation events, even though likely common, remain to be explored. There is considerable evidence of past population isolation - for example, during the quaternary period several European mammals and birds were isolated into glacial refugia, African and non-African Drosophila melanogaster populations were isolated several thousand years ago, and most pathogens and viruses are temporarily isolated within their hosts. Using coalescent theory, we investigate the temporal impact of past population isolation on gene genealogies, on the site frequency spectrum and on summary statistics of DNA sequence polymorphism (e.g. mismatch distributions, Tajima's D, Fay and Wu's H, Zeng *et al.*'s E). We find that past population isolation results in (i) changes in gene genealogies: coalescence trees display an excess of short and long branches, with the relative proportion being informative on the time at which isolation occurred and the number of populations which were isolated. (ii) changes in the site frequency spectrum that vary through time and depend on the sampling scheme. In a sample from a single population, a transient excess of low and high frequency variants is observed, while in a pooled sample from the total population, an excess of intermediate frequency variants and a deficit of high frequency variants is observed. We discuss how the distinctive features of gene genealogies and the site frequency spectrum can be disentangled from other demographic (e.g. bottlenecks) or selective events (e.g. directional, balancing or background selection) and provide recommendations for the analysis of DNA sequence data.

13. Rapid Evolution and Population Genetics

D21SY13PS0975

GENETIC DIVERSITY AND DIFFERENTIATION OF TETRAPLOID PRIMULA HALLERI BASED ON A NOVEL PANEL OF 11 MICROSATELLITE LOCI

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Primula halleri is a tetraploid herbaceous perennial species distributed sparsely in three European mountain chains: the Swiss/Italian Alps, the Carpathians, and the Pirin Mountains (Bulgaria). As one of several polyploid species in Primula subgenus Aleuritia, *P. halleri* represents a model for the loss of the classic Primula distylous reproductive system in response to polyploidy in species endemic to high alpine and arctic environments. It was recently shown that *P. halleri* populations in the Swiss Alps likely exhibit some degree of mixed mating due to a reduction in herkogamy throughout the process of floral development. In order to establish the first estimates of genetic differentiation and diversity in *P. halleri*, we developed a panel of 11 novel microsatellite loci, which we used to genotype specimens sampled from seven populations representing the three main mountain ranges from which this species is known. These data provide the basis of a first assessment of the geographic distribution of genetic diversity in this species, and offer preliminary information regarding the historical processes that may have shaped the distribution of this high alpine endemic. Additionally, our results will provide the first evidence about the origination of the tetraploid nature of *P. halleri*.

RAPID EVOLUTION IN RESPONSE TO CHEMICAL POLLUTION? TESTING FOR THE REQUIRED ADDITIVE GENETIC VARIANCE IN WHITEFISH (SALMONIDAE)

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One of the most common chemical pollutants is 17α -ethinylestradiol (EE2), a synthetic estrogen used in most contraceptive pills. Compared to other estrogens, EE2 is rather persistent in the environment and often found in concentrations that could be harmful to fish development. We set up an experiment (i) to describe the toxicity of EE2 during early development in the whitefish *Coregonus palaea* and (ii) to test whether there is additive genetic variance in the tolerance to the pollutant within a natural population. We sampled a natural population of whitefish and collected their gametes for in vitro fertilization. In order to separate paternal (i.e. additive genetic) from maternal environmental effects on embryo and larval development, we used a full-factorial breeding design, raised large numbers of embryos separately in individual containers, and exposed them to various EE2 concentrations (0ng/L, 0.5ng/L, 1ng/L, 10ng/L and 100ng/L). We are currently recording growth, survival, and gonad development. Variance components can very soon be calculated and will give us an idea about the potential of rapid evolution in a common fish in response to a common pollutant.

A LITTLE SEX GOES A LONG WAY: CLONAL INTERFERENCE DUE TO SPATIAL STRUCTURE CAN BE ALLEVIATED BY LOW RATES OF RECOMBINATION

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The spatial ranges over which populations occur are often much larger than the scales over which individuals migration and interact in a generation, and the resulting spatial structure can have large effects on adapting populations. In particular, the time for a beneficial allele to sweep through the population can be greatly increased if it must spread as a traveling wave over the whole range. It has recently been shown that the resulting increase in clonal interference limits the speed at which spatially-structured asexual populations can adapt, and that this limiting speed is much lower than that of well-mixed population is limited by interference, the limit on the adaptation of spatially-structured populations is not much lower than that on the adaptation of well-mixed ones. Even very low rates of recombination are sufficient to allow spatially-structured populations to adapt at speeds similar to well-mixed ones, suggesting that even organisms that are primarily asexual may be able to adapt fairly rapidly.

EXPERIMENTAL POPULATION GENETICS IN MICE LIVING UNDER SEMI-NATURAL CONDITIONS

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We have established a facility to experimentally study allele-frequency changes in mouse populations living under semi-natural conditions. As a first test-case we have studied the Pldi locus. Pldi is a relatively young gene, which evolved de novo around 2.5 – 3.5 MY ago in the genus *Mus* and is expressed in postmeiotic cells of the testis. Population genetic analysis of this gene region in different subspecies of the house mouse (*Mus musculus*) raised evidence for positive selection at this locus in *M*. *m. musculus*. In order to study the function of this gene, a conditional knockout of the whole gene region has been designed on a C57BL/6 background. Offspring of those knockout mice are viable and fertile, however, a reduction of testis weight and sperm motility could be observed, as well as changes in gene expression of several other genes, which reveals Pldi being part of a regulatory network (Heinen et al. 2009). We have now used semi-natural enclosures with founder populations of 20 mice (10 knockout and 10 wildtype mice) in equal sex ratio each, where mice lived in rooms and could mate freely. Over one year, we monitored all three replicate enclosures regularly every 6-8 weeks and genotyped every mouse in the experiment. In general we aim to answer the question whether we can trace evolution in mouse populations living under semi-natural/semi-controlled conditions. In particular we address the questions whether there are allele frequency changes at the Pldi locus and whether we observe any fitness/reproductive consequences of knockout vs. wildtype mice? Using population genetic analysis tools we can implement all advantages of semi-natural experiments (*i.e.* known demography of those populations and complete sampling of genotypes) to elucidate the evolutionary dynamics and fitness relevance of the Pldi gene.

13. Rapid Evolution and Population Genetics

D21SY13PS1063

GENETIC DIVERSITY AND POPULATION STRUCTURE OF RHIZOPHORA APICULATA, A MANGROVE TREE IN CENTRAL INDO-WEST PACIFIC

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Population genetic analysis of mangroves can reveal their patterns of genetic diversity, which is important for understanding their evolutionary history and scientific conservation. In this study, we examined the nucleotide diversity and population differentiation in six populations of a mangrove tree, Rhizophora apiculata, from China and Thailand based on sequencing 78 nuclear genes through Illumina platform. For each population, more than 30 individuals were pooled for sequencing. The average θ w and θ \pi of the six populations ranged from 0.000263 to 0.000786, and from 0.000258 to 0.000924, respectively, showing low levels of DNA polymorphism within each population. Fst analysis showed strong population structure at the regional level, suggesting Malay Peninsula as a barrier to restrict gene flow between the Andaman Sea and South China Sea despite the existence of the Malacca Strait. However, Fst values vary from zero to one by locus, which could not be explained by simple gene flow model. Haplotype analysis showed two highly divergent haplotypes often reside within populations, which may be caused by recent admixture. Patterns of genetic diversity in this species might result from the repeated glacial events during Plio-Pleistocene, when the Andaman Sea was isolated with South China Sea at the lowered sea level. Reduced genetic diversity in populations from South China Sea can be attributed to lower effective population size for the enormously changed shorelines during this stage. We further used Sanger method to obtain sequences of three genes among 15 populations from wider geographic regions. STRUCTRUE analysis indicated that these populations formed five clusters, corresponding to five geographic regions, which suggests population structure of this species was mainly shaped by geographic isolation. Our study reveals the effect of glacial events on patterns of genetic diversity of an intertidal mangrove tree, which will be informative for its conservation.

MUTATION AND MIGRATION: WHEN AND WHERE DOES DIVERSITY ARISE IN EARLY SIV INFECTION?

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Introduction The rapid evolution of HIV plays a role in disease progression, drug resistance evolution, and hinders vaccine development. The early stage of infection is of particular importance as the virus populates a tenacious reservoir of latently infected cells, preventing clearance of infection. There is evidence for isolation of HIV sequences between different tissue types, but where sequence diversity arises, how fast it spreads and to where, is poorly understood. This is largely because the first weeks of infection are almost unstudied due to the low detectability of virus, and uncertainty as to the timing of natural infections. Methods We explore this early stage in next generation (454) sequencing data from 15 Rhesus macaques experimentally infected with SIV, as a human model for HIV infection. Virus was sampled at multiple time points (7 to 168 days) and tissue compartments (lymph node, rectum, blood cells and plasma). We examine the change in diversity between compartments over time, and where particular mutations arise. By fitting simple and more complex models we estimate the rates of migration between body compartments. Results Diversity generally increases over time, but often falls later in infection, which corresponds to the rise of escape mutations. In agreement with previous analysis of this dataset, escape mutations tend to occur first in the lymph nodes. The migration rate from the gut tissue into the lymph nodes is higher overall than in the other direction. However the migration rates between body compartments are highly variable between animals, which may reflect either stochastic effects or host-based differences in the course of infection. Conclusion Rich data from next generation sequencing provides many opportunities to study spatial evolution in early immunodeficiency virus infection. These data present challenges such as a high error rate, but careful modeling and treatment of the assumptions compensates for such issues.

INFLUENCE OF TEMPORAL VARIABILITY OF MATING SYSTEM ON THE ADAPTATION PROCESS AFTER AN ENVIRONMENTAL CHANGE

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The young age of selfing lineages is often interpreted as evidence that selfing species lose the ability to adapt to changing environments. Besides influencing genetic structure or interacting with life history traits, mating system can constrain the adaptation process. In one hand, self-fertilization leads to an increase of genome-wide homozygosity, which exposes recessive beneficial mutations to selection. On the other hand, it also reduces the effective size, which in turns decreases the efficiency of selection with respect to drift, and ultimately reduces standing variation. Consequently, mating system should strongly influence adaptation. Recently, mixed mating strategies have been shown to occur at a substantial frequency, especially among animal-pollinated taxa. In species with mixed mating system, the selfing rate is extremely variable and potentially plastic both between years, environmental conditions, populations and families. Several ecological factors, such as plant density or pollination ecology may influence the relative importance of self-fertilization and outcrossing in hermaphrodite plants. Variation of these environmental factors may yield rapid variation in plant mating system, which consequences on the process of adaptation have been little explored. The effect of temporal variability of mating system is explored using simulations in order to better understand its consequences on the probability and speed of adaptation for a population facing environmental change. We model the fixation dynamics of a beneficial mutation, varying its dominance, its time of appearance, and the pattern of temporal variation in selfing rates. We focus on mainly outcrossing populations to test whether occasional events of selfing could maximize the rate of adaptation by favouring the expression of beneficial recessive mutations while not compromising the maintenance of standing variation.

POPULATION GENETICS OF COPY NUMBER VARIABLE GENES IN NATURAL MOUSE POPULATIONS

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Copy number variation (CNV) is a form of genomic structural changes that are considered a major source of intraspecific genetic variation. It is not unusual that CNVs encompass entire genes and their regulatory sequences. As differences in gene copy number can underlie fitness differences between individuals, studying the population dynamics of copy-number variable genes can provide valuable clues to selection and adaptation processes. Here, we analyze genic CNVs from next-generation sequencing data of 27 individuals belonging to four natural populations of the house mouse (*Mus musculus*). We find that the environmental response genes are overrepresented in genic CNVs in all four populations. CNVnator software was used to genotype all the loci for their respective copy number. This allowed us to examine their variance within and across populations. Apart from the strong population stratification in geographically distinct groups, we observe much higher level of diversity between mouse populations than was previously reported for human populations. Several genes were identified that show no CNV within some populations while being variable in others. These cases stand out from a generally polymorphic CNV background, implying that at least some of them might be subject to selective constraints.

13. Rapid Evolution and Population Genetics

D21SY13PS1268

INFERRING THE INVASION ROUTES OF AN ANCIENT INVADER, THE MEALYBUG PEST SPECIES *PSEUDOCOCCUS VIBURNI* SIGNORET (*HEMIPTERA: PSEUDOCOCCIDAE*), BY USING MICROSATELLITE MARKERS AND DNA SEQUENCES

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To infer population invasion history, programs based on approximate Bayesian computation (ABC) have proved useful to infer the most probable invasion scenarios in a range of biological systems.
br>Pseudococcus viburni is a cosmopolite agricultural pest, probably of South American origin. Data suggests that P. viburni distribution expansion started around 1600 with first potatoes trades and quarantine reports starts around 1890. In terms of population genetics, P. viburni population structure is composed of many small populations displaying very little allelic richness, i.e. populations that have probably all undergone severe demographic bottlenecks even in the native area. This ancient invasion history and this particular population genetic structure make P. viburni an interesting and challenging case for the inference of population worldwide history, with multiple possible invasion pathways between all the continents. In order to characterize precisely the population genetic structure of the species at worldwide scale, and infer its invasion history, we collected 30 populations and genotyped them using 25 microsatellite markers. Summary statistics showed a remarkable population differentiation, with Fst ranging from 0 to 0.8. These values can be explained by P. viburni low mobility, its dispersal being mainly driven by human activities. To infer the invasion history the DIYABC program was used with microsatellite and sequence data. An ancient bottleneck of around 460 generations (~150 years) was inferred. This event fit with first species reports. Then several introduction scenarios were tested to infer the invasion routes of P. viburni to Europe and North America. In this work we discuss the origin and invasion routes for P. viburni by comparing the results obtained from different methodologies: traditional statistics, assignment tests and ABC methods based on parameters estimated fom the data generated with different types of genetic markers.

DIVERGENCE IN GENE EXPRESSION IS UNCOUPLED FROM DIVERGENCE IN CODING SEQUENCE DURING RAPID EVOLUTION IN HELIANTHUS

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The transition from herbaceous annual to woody perennial has occurred numerous times during the evolution of flowering plants, but the genomic changes that accompany this ecological transition remain poorly understood. Here we present an analysis of genetic sequence and gene expression divergence between the annual *Helianthus annuus* and the recently diverged, woody perennial species *H. winterii* from California. This new species has likely diverged within the last several thousand years, and during this period has rapidly adapted to a novel, marginal habitat in the Sierra Nevada foothills. *Helianthus winterii* flowers later and for a longer period, and is taller at maturity than typical *H. annuus*. Using flow cytometry we confirmed that the new species has a genome size on par with typical *H. annuus*, and therefore major genomic duplication is an unlikely explanation for the ecological transition. We sequenced the transcriptomes of six individuals of each species and discovered 575 genes (3.5%) with significantly different expression between the species, as well as seven biological processes associated with divergent gene expression. Surprisingly, only five of the differentially expressed genes were also present among genes with the most coding sequence divergence (top 3.5% of FST distribution). These two mechanisms may represent alternate pathways towards the same adaptive optimum, or genes may be alternatively constrained over short time scales.

OCCUPATION OF A NOVEL ENVIRONMENTAL NICHE VIA HYBRIDIZATION IN CENTRAL ASIAN WATER FROGS PELOPHYLAX

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Many animal groups inhabiting Central Asia (CA) have complex biogeography, shaped by Plio-Pleistocenic climate and geological events. Relatively recent aridization makes this region interesting from evolutionary perspective as it represents an ideal arena to study evolution of aquatic species as water frogs *Pelophylax*. Until recently, this genus was considered to be represented by a single species in CA, however our phylogenetic analysis of two mitochondrial and one nuclear loci revealed presence of two divergent water frog taxa: P. terentievi and P. sp., each inhabiting a distinct river drainage. Sympartic occurrence of the heterospecific mtDNA haplotypes in the interflow of these two rivers and presence of heterozygots for the nDNA locus pointed on existence of a complex hybrid zone. Subsequent analyses of 8 codominant loci with STRUCTURE and TESS confirmed presence of 2 distinct genotypic clusters (Fst=0.48) as well as area of their admixture. Inference of admixture classes (NEWHYBRIDS) and the cline analysis (CFit7) pointed on tension type of the hybrid zone. Further, the TESS spatial interpolation map combined with the mtDNA haplotypes distribution data pointed on a zone of unidirectional mitochondrial introgression from *P. terentievi* to the *P.* sp. population. We applied environmental niche modelling (MaxEnt) to verify how different are parental populations (n=24 of each *P. terentievi* and *P.* sp.) and ones with mt introgression (n=5), having used 19 bioclimatic and 13 other layers. The obtained prediction models were highly reliable (mean AUC=0.997). Mean winter temperature had unique and high (accounted for 36% of the predicted range) impact on the model of populations with introgressed mitochondrial DNA. Hence, we speculate that hybridization of the two divergent water frog species and subsequent introgression of heterospecific mitochondrial DNA into the marginal populations of *P*. sp. promoted them to occupy novel ecological niche.

13. Rapid Evolution and Population Genetics

D21SY13PS1284

EVOLUTIONARY BIOLOGY OF THE INVASIVE NORTHERN PACIFIC SEASTAR (ASTERIAS AMURENSIS) IN AUSTRALIA

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Biological invasions present interesting evolutionary problems because they are often stochastic events often involving small populations that must survive and adapt to novel habitats. Thus, invasive species offer rich opportunities to investigate the effects of reduced diversity and population bottlenecks on the rate and predictability of evolution in the wild. One of the most successful invaders into Australian waters over the last decades is the predatory Northern Pacific Sea Star (*Asterias amurensis*). The source population is believed to be from Japan and was introduced into Australia via ballast water discharge, although it remains unclear if multiple introductions have occurred. Here we present the results of a genetic survey of the introduced Australian populations and compare it to populations in it native range in Japan. The potential for this species to increase its geographical range in Australian waters and its potential genetic limitations are discussed.

LINKAGE-BASED METHOD FOR IDENTIFYING SELECTED LOCI AND CHARACTERISING SELECTION INTENSITY

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A critical element in our understanding of selection is the ability to detect and characterise it using genetic data. Recently, several linkage disequilibrium based methods have been developed and widely applied to identify selective sweeps in human populations. However, methods to determine the intensity of selection and the time that selection started and the time that it ceased, are generally still underexplored. This is because teasing apart the impact of these factors on the summary statistics affected by selection is often complex. We have developed a method for identifying fixed and high frequency selected loci, and characterising selection intensity, that attempts to control for the effects of evolution in the period since selection ceased. The approach follows others in focussing on the differing linkage patterns along the chromosome on either side of a positively selected locus as compared to those over the locus. During selection, the impact of recombination on these regions is known to be different, while after selection has ceased the linkage decay becomes similar. Although methods of assessing selection intensity exist based on diversity and haplotype frequency spectra, controlling for post-selection evolution adds subtlety to our understanding of the process. We have characterised the performance of this approach and tested it using numerical methods and coalescent simulations.

13. Rapid Evolution and Population Genetics

D21SY13PS1358

POST-INTRODUCTION EVOLUTIONARY RESCUE

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Invasive mosquitoes have caused inordinate human suffering because they are very abundant in their exotic ranges and can drive local or exotic disease evolution and epidemics. Invasive mosquitoes often exploit domestic environments, and the resulting extensive overlap with humans significantly enhances their effectiveness as vectors of human diseases. It is unclear, however, whether domestication was a pre-condition for invasiveness or instead a consequence of it. To understand factors that deter or promote invasiveness we studied Aedes japonicus japonicus, a mosquito native to East Asia that has become invasive in some parts of its new range in North America and Europe. We traced the source of all exotic populations to central Japan, where we found high genetic diversity but homogeneity across populations indicating ongoing gene flow. Spatiotemporal analyses of exotic populations revealed that (1) populations across the exotic range had very different genetic signatures; (2) genetic diversity declined precipitously outward from introduction points; (3) the spatial extent of local infestations correlated strongly with genetic diversity but at different scales in the US and in Europe; (4) all broadly expanding populations (=invasive) had genetic signatures resulting from admixture of separate introductions. We propose that human-assisted rapid movement of specimens across the exotic range "rescued" self-limiting introductions by mixing locally differentiated genotypes. We propose that invasiveness is not a required pre-existing trait in invasive mosquitoes, but instead can evolve in the exotic range leading inexorably to dangerous human disease vectors.

ADAPTIVE EVOLUTION OF THE OPSIN GENES IN AFRICAN CRATER LAKE CICHLIDS?

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Cichlid fishes from volcanic crater lakes represent small radiating lineages usually of few species. The Barombi Mbo Lake in Cameroon hosts a species flock of tilapiine cichlids, which underwent adaptive radiation similar to more famous Great Lakes cichlids. Several ecological adaptations were observed either in morphology of trophic apparatus (leading to feeding specializations) or extreme habitat preferences, such as deep-water niche. In general, cichlids are mainly visually oriented animals and their visual system shows in many cases the additional adaptations to the environment including the deep waters. Two ways of adaptation were found in cichlids from the Great Lakes, first in the DNAsequence tuning and second in the gene expression levels differenciation. In the deep-water species convergently evolved mutations are known in opsin genes causing the shift of the chromophore sensitivity. The Barombi lake hosts a deep-water living species Konia dikume, however, nothing is known about the opsin evolution in these small crater lake systems so far. We sequenced a large region (up to 10 kbp) of each of the seven cone opsins (and putative regulatory regions) using the amplicon sequencing in the Ion Torrent next-generation sequencer. We further performed whole-transcriptome sequencing on the Ion Torrent next-generation sequencer to reconstruct robust phylogeny and we reconstruct evolution of the opsin genes by mapping on the tree. We further estimated the expression levels of all seven cone opsin genes, as well as the rod opsin. We found faster mutation rate in opsin genes in the genus Stomatepia (three species) and higher allelic variation within the genus Sarotherodon, however no DNA tuning has been found in Konia dikume.

INFERRING CONSERVATION AND ADAPTATION OF MOLECULAR QUANTITATIVE TRAITS

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Molecular phenotypes are important links between genomic information and organismic functions, fitness, and evolution. Quantitative traits, such as gene expression levels, depend on multiple genomic loci, and their evolution involves selection, genetic drift, mutation and recombination. In this talk, we discuss a new method to infer conservation and adaptive evolution of such traits. The method is based on the trait divergence between populations in a phylogeny. This statistics decouples from details of the defining genotypes. In particular, we consider evolution in a single-peak fitness seascape, which defines optimal trait values changing over evolutionary time. Our method infers both the strength and the time-dependence of the fitness seascape. We discuss the implications of our findings for the predictability of evolutionary processes.

Symposium

14. Non Genetic Inheritance

22 and 23 August



Program

Thursday 22 August

Session(s): 9 Friday 23 August

Session(s): 10, 11

Organisers: Benoit Pujol and Katie Stopher

Invited speakers: Kevin Laland and Etienne Danchin

Description:

Evidence is accruing that epigenetic, developmental, parental, ecological and cultural inheritance mechanisms have a major impact on the evolution of phenotypic diversity. The aim of this symposium is to highlight novel results and synthesize our knowledge on the contribution of non-genetic inheritance to evolutionary processes. We will also explore the need for an extended theory of evolution where genes are not the only inheritance system.

14. Non Genetic Inheritance

D22SY14IT10:30R6

GENETIC AND NONGENETIC INHERITANCE

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Many biologists are calling for an 'extended evolutionary synthesis' that would 'modernize the modern synthesis' of evolution. Biological information is typically considered as being transmitted across generations by the DNA sequence alone, but accumulating evidence indicates that both genetic and non-genetic inheritance and their interactions have important effects on evolutionary outcomes. I will define the terms of nongenetic inheritance and briefly review the evidence for such effects of epigenetic and cultural inheritance, and outline methods that quantify the relative contributions of genetic and non-genetic heritability to the transmission of phenotypic variation across generations. These issues have major implications for diverse domains, including medicine where they may profoundly affect research strategies. For instance, non-genetic inheritance may explain a significant part of one of the major enigma of current molecular biology, namely missing heritability, which concerns many human supposedly genetic disorders. The missing heritability rather suggests that we should broaden our view of inheritance by including non-genetic inheritance into an inclusive theory of evolution.

D23SY14IT14:00R6

GENETIC CONTROL OF NON-GENETIC INHERITANCE: FACT OR FALLACY?

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The foundation of the Modern Synthesis placed the gene at the centre of evolutionary explanations of biology, largely because at the time Mendelian genetics appeared to be the only general inheritance system. More recently, other non-genetic inheritance (henceforth NGI) systems (epigenetic inheritance, parental effects, ecological inheritance, social transmission and cultural inheritance) have been identified and been found to be widespread, raising questions about whether, and how, non-genetic inheritance might change the character of biological evolution. Here we draw on established empirical and theoretical findings from the cultural evolution, gene-culture coevolution, evolution of plasticity, and niche construction, literatures to illustrate ten ways in which non-genetic inheritance affects biological evolution. These include the findings that NGI systems can generate non-random (adaptive) variants; that they change the rate and dynamics of evolution, the pathways of information flow, and equilibria reached; that they alter the frequencies and spatial distribution of phenotypes; that they can propagate maladaptive variants; and that they change conceptions of fitness. These findings leave untenable the claims that non-genetic inheritance is under genetic control and as a consequence does not change the evolutionary process in biologically meaningful ways. Our analysis supports arguments for a broadening of current conceptions of biological evolution.

D22SY14RT11:18R6

THE INFORMATION VALUE OF NON-GENETIC INHERITANCE IN HETEROGENEOUS ENVIRONMENTS

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Parents contribute a variety of inputs to the development and fitness of their offspring beyond the transmission of DNA, including epigenetic marks, transfer of nutrients, antibodies and hormones, and behavioural interactions after birth. The evolutionary consequences of such non-genetic inheritance have been explored recently. By disconnecting what is selected from what is inherited, non-genetic inheritance can modify the course of evolution and selection on future generations. Less is known, however, about how mechanisms of non-genetic inheritance have themselves evolved. Here, we present a simple model to explore the adaptive evolution of non-genetic inheritance under different regimes of environmental change. Our model is based on a developmental switch that can evolve to produce different phenotypes in response to different levels of input. We consider genetic and nongenetic inputs as potential cues containing correlational information about coming selective conditions. Differential use of these cues is manifested as different degrees of genetic, parental or environmental morph determination. By exploring a range of temporal and spatial environmental fluctuation scenarios (cyclic and stochastic, of varying frequency), we evaluate the conditions that favour non-genetic inheritance as opposed to genetic determination of phenotype or within-generation plasticity. Finally, we use the model to exemplify three case studies which have provided hallmark examples of nongenetic inheritance: maternal effects on seed germination in plants, transgenerational phase shift in desert locusts and grandparental effects on dispersal polymorphisms in aphids.

D22SY14RT11:42R6

NON-GENETIC INHERITANCE GENERATED BY EXPOSURE TO ANTHROPOGENIC STRESSORS IN THE WATER FLEA (DAPHNIA PULEX)

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Understanding how populations rapidly adapt to anthropogenic change is imperative for predicting limits to population persistence and reducing species extinction rates. Since most random mutations decrease fitness, and even those that increase fitness initially only occur in a single individual in the population, it is hard to envisage how new mutations alone can explain rapid evolutionary responses. Rapid adaptation might arise through 'soft selection' operating on standing genetic variation. However, an emerging, alternative hypothesis that is that environment-induced non-genetic inheritance facilitates and speeds up adaptive evolution.
by decoupling phenotypic change from genotypic change, nongenetic inheritance (parental effects and epiallelic variation) is not subject to the limitations typically associated with genetic inheritance. However, our lack of understanding of the mechanism that underpin non-genetic inheritance, the transience and instability of non-genetically transmitted phenotypic states, and the way that non-genetic inheritance interacts with genetic inheritance, all greatly limit our ability to evaluate the significance non-genetic inheritance for long-term evolutionary change. We addressed this shortfall by performing a 4-generation experiment in which we exposed 3 clones of *Daphnia pulex* to sub-lethal doses of novel freshwater pollutants (heavy-metal, endocrine disruptor, herbicide) and then quantified the patterns of non-genetic inheritance generated over the next three generations. This was done at the individual trait level as well as at the multivariate level, using phenotypic trajectory analysis to quantify stressor-induced patterns of phenotypic integration over multiple generations. We evaluate whether non-genetic inheritance accumulates, persists or dissipates over multiple generations, whether these patterns differ for different types of stressor, and whether there is an interaction between genetic and non-genetic inheritance.

D23SY14RT10:30R6

MATERNAL EFFECTS ON OFFSPRING GROWTH, SURVIVAL AND SOCIAL STATUS IN SPOTTED HYENAS

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We have studied two processes of non-genetic inheritance in a social mammal with linear dominance hierarchies. Firstly, in such species, the social status of an offspring at adulthood is often similar to the social position held by their mother; a phenomenon termed "rank inheritance". Mothers may influence the rank obtained by their offspring at adulthood in at least three distinct ways: 1) the direct genetic inheritance of maternal traits that influence resource holding potential might predispose offspring to obtain a rank similar to that held by their mother, 2) the pre-natal maternal environment might influence offspring rank if foetal exposure to maternal androgens is related to maternal status and affects offspring competitiveness, and 3) maternal behavioural support, a component of the post-natal maternal environment, may help offspring to dominate those individuals that are subordinate to their mother, thereby assisting offspring to acquire a rank similar to that of their mother. We simultaneously test predictions derived from these three potential maternal effects on offspring rank acquisition at adulthood, using rare cases of offspring adoption detected by microsatellite profiling. We demonstrate that adopted offspring acquire a rank at adulthood similar to that of their surrogate mother and that the competitive ability of offspring at adulthood was best explained by post-natal maternal behavioural support. Secondly, we use long-term data to also show that high-born offspring have higher growth rates, are more likely to survive to adulthood and start reproduction at an earlier age than offspring of lower ranking mothers – thereby demonstrating a maternal 'silver spoon effect' in spotted hyenas.

D23SY14RT10:54R6

PATERNAL EFFECTS ON LIFESPAN AND AGING IN THE FRUIT FLY (DROSOPHILA MELANOGASTER)

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Paternal effects are a potentially important source of non-genetic inheritance; however, attempts to quantify natural variation in paternal effects are often complicated by or confounded with maternal effects or the effects of genetic variation. Here we present a novel method for isolating and quantifying paternal effects using hemiclonal analysis. In *Drosophila melanogaster*, hemiclones have been used to replicate an entire haploid genome while holding the other haploid genome constant. We created hemiclones from diverse genetic backgrounds, including 38 inbred lines from the Drosophila Genome Reference Panel and 4 lines derived from other global populations. By crossing these hemiclones to homozygous females, we can select flies that are genetically identical, despite having fathers from genetically diverse backgrounds. We screen these 42 lines for variation in lifespan, a phenotype that harbors significant variation in the source populations, and is affected by a wide variety of genetic and non-genetic factors. Preliminary results indicate that there are significant differences in lifespan attributable to variation in paternal effects, and these results will further explore the relative importance of standing variation that exists within populations compared to the divergent variation that exists between populations. We will also consider the relative importance of paternal effects on early- vs late-stage mortality.

D23SY14RT11:18R6

THE FITNESS IMPLICATIONS OF ADAPTATION VIA PHENOTYPIC PLASTICITY AND MATERNAL EFFECTS

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In a changing environment, organisms face the challenge of using the most accurate environmental cues to construct phenotypes that optimise fitness. This environmentally specific phenotypic expression is delivered by phenotypic plasticity within the current generation and potentially also via previous generations through indirect or nongenetic inheritance. Using a quantitative genetic model of adaptation via indirect genetic maternal effects, phenotypic plasticity and an additive genetic component, we show how the maternal effect coefficient that maximises population mean fitness depends on the predictability of environmental change (fast or slow, sinusoidal or stochastic) and the lag between development and selection. The impact of this developmental lag is particularly clear in rapidly changing stochastic environments. We consider the influence on fitness of interactions between phenotypic plasticity and maternal effects. Expected mean population fitness is highest away from the peak levels of phenotypic plasticity, particularly in slowly or rarely changing environments. We expect the relative influence of phenotypic plasticity and maternal effects to differ among environments, which may help to explain why we observe such a vast range of maternal effects coefficients empirically.

D23SY14RT11:42R6

LIKE FATHER LIKE SON? NONGENETIC PATERNAL EFFECTS REINVIGORATE THE POSSIBILITY OF TELEGONY

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Telegony, a belief that was once widely accepted but now largely dismissed, is the hypothesis that offspring occasionally inherit characters from a previous mate of their mother. Although telegony seems unfeasible under Mendelian laws of inheritance, mounting empirical evidence for nongenetic inheritance mechanisms reinvigorates the hypothesis. In the neriid fly, *Telostylinus angusticollis*, fathers transmit their environmentally-acquired condition to offspring: large fathers that are reared on a high-quality larval diet produce larger offspring. Males show no obvious forms of paternal investment or care, and thus the mechanism mediating the effect of paternal condition on offspring body size is not known. As a first step towards disentangling whether the effect is borne by the sperm itself or by accessory-gland products (ACPs) in the seminal fluid, we mated females initially to a male in high or low condition two weeks later. Interestingly, offspring size and viability were determined by the condition of the first male, with no effect of the condition of the second mate. Genetic tests confirm this result holds even when the second male is the biological father of the offspring. These findings suggest the paternal effect is mediated by ACPs, and provide a compelling case for reassessing the possibility of telegony as a valid phenomenon.

D23SY14RT14:48R6

SEX SPECIFIC SOCIAL GENETIC EFFECTS IN PARENTAL-CARE BEHAVIOUR

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The social environment, namely conspecifics, can influence the expression of phenotypes i.e., individuals other than the one expressing the trait in question can explain some of the phenotypic variation. The genetic part of this indirect effect represents an indirect genetic effect (IGE). Not accounting for IGEs can considerably under- or over-estimate the total heritable variation available for selection to act on, and thus predict misleading evolutionary trajectories. Yet, empirical studies on wild populations often ignore IGEs. We present a quantitative genetic analysis of biparental care in a wild, genetically pedigreed, bird population. Sex-specific IGEs increased the total heritable variation. Our data suggests that the female trait could evolve through indirect selection by her mating partner, which most theoretical models explaining the evolution of biparental care do not take into account. Notably, the within-individual repeatability of female parental care was lower than the total heritable variation. Our results show that the assumption that repeatability is the upper limit of heritability should be used cautiously when applying to socially interactive traits, and highlight the importance of accounting for social effects.

D23SY14RT15:12R6

THE RELATIVE IMPORTANCE OF GENETIC AND NONGENETIC INHERITANCE IN RELATION TO TRAIT PLASTICITY

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A trait's response to natural selection will reflect the nature of the inheritance mechanisms that mediate the transmission of variation across generations. The relative importance of genetic and nongenetic mechanisms of inheritance is predicted to be related to the degree of trait plasticity, with nongenetic inheritance playing a greater role in the cross-generational transmission of more plastic traits. However, this prediction has never been tested. We investigated the influence of genetic effects and nongenetic parental effects in two morphological traits differing in degree of plasticity by manipulating larval diet quality within a cross-generational split-brood experiment using the seed beetle *Callososbuchus maculatus*. In line with predictions, we found that the more plastic trait (elytron length) is strongly influenced by both maternal and paternal effects whereas genetic variance is undetectable. In contrast, the less plastic trait (first abdominal sternite length) is not influenced by parental effects but exhibits abundant genetic variance. Our findings support the hypothesis that environment-dependent parental effects may play a particularly important role in highly plastic traits and thereby affect the evolutionary response of such traits.

POSTERS

THE MOLECULAR BASIS OF ADAPTIVE MATERNAL EFFECTS IN AMPHIBIANS

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Maternal effects, the effect of a mother's phenotype and environment on offspring phenotype and performance, can influence the speed and direction of trait evolution, responses to divergent selection, as well as population dynamics. However, the mechanisms and the molecular basis of maternal effects are often poorly understood. Here we present data for natural populations of two amphibian species (*Rana arvalis* and *Rana temporaria*), which show intra-specific adaptive divergence to acidity via maternally derived egg capsules. Particularly, we use cutting edge molecular approaches (proteomics, glycan analysis) to elucidate the molecular mechanisms behind adaptive maternal effects in these systems, and emphasize the role of egg capsules for adaptation at early life-stages.

(NON)-GENETIC INHERITANCE: A HISTORICAL OVERVIEW

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Current debates on the biological relevance and theoretical impacts of non-genetic inheritance in evolution seem to have much in common with archaistic debates between Neo-Lamarckians and Neo-Darwinians on the existence of inheritance of acquired characteristics. In this presentation, we will adopt a historical point of view to put the current debates into perspective. In the first part, we will first recall how the publication of The Origin made the researches about inheritance a central theme of theoretical biology, and how these researches themselves put Darwinism at risk at the turn of the XIXth-XXth centuries. We will recall how Neo-Darwinism and Neo-Lamarckism emerged during the debates on the role of acquired characteristics in heredity. We will show how, as soon as the end of the XIXth century, the inheritance of acquired characteristic has been a heterogeneous concept – heterogeneous enough to seem, in some instances, compatible with Neo-Darwinism. In the second part, we will show how, in the first part of the XXth century, researches on genetic inheritance impacted the debates about the inheritance of acquired characteristics, making them slip towards debates on the "genetic" inheritance of acquired characteristics. We will show how, in parallel, researches on nongenetic inheritance got marginalized, without disappearing totally, before blossoming again at the end of the XXth century. In conclusion, we will draw on our historical account to clarify the conceptual differences between directed mutation, the inheritance of acquired characteristics, and non-genetic inheritance. This will lead us to consider in which respect an Extended Evolutionary Theory could depart from the conceptual matrix of the Modern Synthesis.

THE BENEFITS, COSTS AND EVOLUTIONARY CONSEQUENCES OF GENOMIC IMPRINTING

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Under genomic imprinting, alleles "remember" which parent they are derived from and show differential gene expression. Imprinting is an evolutionary puzzle because it carries all the costs of diploidy but foregoes its principal benefit (protection from recessive mutations). Here, I synthesise and critically evaluate the many evolutionary hypotheses for the origin and maintenance of imprinting, while adding at least one more. I also review imprinting's many costs, and argue that the origin of imprinting is substantially more difficult than the maintenance because of evolutionary changes wrought by imprinting itself. I finish by using theoretical models to consider the long-term consequences of imprinting for adaptation and population fitness.

MATERNAL EXPOSURE TO PREDATORS: HOW TO PREPARE OFFSPRING TO A RISKY NATAL ENVIRONMENT?

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Predation risk is a strong force inducing morphological, physiological and behavioural responses. As predation risk is often higher in early life, antipredator defences should be produced at a juvenile stage. Maternal effects are a good tool to produce antipredator defences early in juvenile life. We investigated if maternal exposure to predator cues during gestation affected juvenile morphology, behaviour at birth and life-history traits in common lizard (Zootoca vivipara). We exposed 22 adult females to cues from a saurophagous snake (*Hierophis viridiflavus*) for one month of gestation and 22 other females were kept unexposed. At birth, juveniles were measured and tested for thermal preferences and activity levels in presence or in absence of predator cues, and then released in semi-natural enclosures connected to a corridor allowing to monitor dispersal. We also quantified survival and growth rate at the end of the season by capture-recapture sessions. Offspring born from exposed mothers had longer tails at birth and at an older age and preferred lower temperatures than juveniles from unexposed mothers. Tail autotomy and decreased basking behaviour are common antipredator behaviours and thus our results suggest that mothers can prepare juveniles to risky environments. Juveniles from exposed mothers also increased their activity levels in response predator cues showing better abilities to recognize such cues. This increased activity, along with an overall increase in dispersal probability, show that mother also manipulated juvenile flight response. In response to predation risk, mothers can manipulate offspring phenotype to make them more adapted to their natal environment or increase their dispersal ability to find more suitable habitats. Our results show that, in common lizards, these two adaptive mechanisms act conjointly in order to improve offspring survival to predators.

DIFFUSION OF SOCIAL INFORMATION WITHIN DROSOPHILA GROUP

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Understanding how behavioral diversity arises and is maintained is central to evolutionary biology. Genetically based inheritance has been a predominant research focus of the last century; however, nongenetic inheritance, such as social transmission, has become a topic of increasing interest. How social information impacts behavior depends on the balance between information gathered directly through personal experience versus that gleaned through social interactions and on the diffusion of this information within groups. We investigate how female *Drosophila melanogaster* use social information under seminatural conditions and whether this information can spread and be maintained within a group, a prerequisite for establishing behavioral transmission. We show that oviposition site choice is heavily influenced by previous social interactions. Naïve observer flies develop a preference for the same egg-laying medium as experienced demonstrator flies conditioned to avoid one of two equally rewarding media. Surprisingly, oviposition site preference was socially transmitted from demonstrators to observer flies had previous personal experience with both rewarding media. Our findings shed light on the diffusion process of social information within groups, on its maintenance, and ultimately, on the roots of behavioral local adaptation.

TRANSGENERATIONAL PLASTICITY IN MARINE STICKLEBACKS: MATERNAL EFFECTS MEDIATE IMPACTS OF A WARMING OCEAN

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Transgenerational plasticity is a potentially powerful mechanism for species to cope with rapid environmental change. Recent studies show these non-genetic parental effects can facilitate acclimation to environmental stressors (e.g. increased temperature and ocean acidification) across generations. Yet, the relative contribution of maternal vs. paternal effects is not well known. We crossed adult marine sticklebacks acclimated to two experimental temperatures (17°C and 21°C) to produce egg clutches in 4 groups: 17mx17f, 17mx21f, 21mx17f, 21mx21f. Clutches were split and reared at 17°C and 21°C. Egg traits (clutch size, egg size and hatching success) and offspring growth at 30 and 60 days were quantified. We found that 21°C females produced smaller (but not fewer) eggs with a lower hatching success than 17°C females, suggesting that mothers optimise their fitness over that of their offspring in stressful environments. Still, offspring growth at 30 days showed a clear benefit of transgenerational (maternal) plasticity; offspring had higher growth when reared at their maternal temperature. Growth at 60 days was mostly determined by offspring environment, yet maternal effects persisted in the stressful environment. Our results show that transgenerational plasticity can mediate some of the impacts of a warming ocean and may facilitate population persistence under climate change.

LARVAL DIET ALTERS ASSOCIATIONS WITH COMMENSAL GUT BACTERIAL AND INDUCES TRANS-GENERATIONAL EFFECTS ON HOST FITNESS IN CERATITIS CAPITATA

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The importance of diet and gut bacteria for host health and fitness is well known across vertebrates and invertebrates. In many instances these associations are variable and flexible, but still of fundamental importance to our understanding of animal biology. In the Mediterranean Fruit Fly (medfly) a pest insect of great agricultural importance, it is known that interactions between diet and gut bacteria can have a significant influence on host development, female fecundity, male mating speed and female survival. There can even be vertical transmission of commensal gut bacteria from mother to offspring, via inoculation into the fruit during oviposition and subsequent ingestion by the developing larval offspring. The composition of microbes is affected by larval and adult diet, which explains why laboratory-reared fruit flies, with a relatively invariant diet, generally seem to have restricted microbial communities in comparison to wild fruit flies. However the precise relationship between diet, gut bacteria and host fitness has not been systematically studied in this pest of economic importance. We address this gap by a study in which we systematically altered larval dietary components and found effects on host development, mating success and gut microbiota. In addition we present data to show that these were trans-generational effects, with non-genetic parental influence on the development of offspring.

SEX DETERMINATION IN THE HAPLODIPLOID ANT SPECIES CARDIOCONDYLA OBSCURIOR

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Sex determination across insects seems to rely on the same pathway: A primary signal induces sex specific alternative splicing of *transformer* (*tra*) or its ortholog. This leads to a functional TRA protein in females, whereas an in-frame stop codon in the male transcript leads to no functional protein. TRA induces female specific splicing of *doublesex* (*dsx*) and the sex specific DSX protein regulates downstream development of the embryo. While the developmental cascade is conserved across taxa, the primary inducing signals are not, ranging from a single locus (*csd*) in *Apis* to maternal imprinting in *Nasonia* (reviewed in Verhulst et al., 2010). We investigated the sex determination pathway in *Cardiocondyla obscurior* (Hymenoptera, Formicidae), a model for eusocial traits. Single locus sex determination can be ruled out, because no diploid males were detected despite many generations of inbreeding (Schrempf et al., 2006). We confirmed the exclusive maternal origin of males by genotyping F1-hybrid offspring of a cross of two distinct populations with known genotypes. The sex determination genes *tra* and *dsx* were annotated and the cDNA sequences were compared between female and male embryos. Furthermore *tra* expression in fertilized versus unfertilized eggs was analysed using RT-qPCR to test for maternal provisioning of the zygote as an epigenetic effect and as the primary signal of the sex determination pathway in *C. obscurior*.

MATCHING OR MISMATCHING: EFFECTS OF MATERNAL HEAT STRESS ARE MODIFIED BY POSTNATAL HEAT STRESS IN JAPANESE QUAILS

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The environment can influence an animal phenotype during various stages of an animal's life but during embryonic development animals are especially susceptible to changes in their environment due to the speed and complexity of development at this time in life. Changes in the prenatal environment often reflect and are due to changes in the parent's environment. Phenotypic alterations induced during this early period in life are therefore particularly intriguing because they demonstrate that environmental changes in one generation can influence the development and phenotypic expression in the next generation. Currently researchers are exploring the role of maternal stress during egg formation in oviparous species as an inducer of phenotypic plasticity in offspring. Maternal stress is often reported to reduce the phenotypic quality of the offspring, but it has also been suggested that these maternal effects might prepare offspring for a stressful environment. However, in most studies the environmental conditions in the postnatal environment in which the offspring are raised and tested are ignored. In this study Japanese quail females and their offspring were either heat stressed (35 °C) or housed at control temperature (22 °C) in a split-brood design. Offspring matched to their mother's hot environment had a significantly faster corticosterone response to 10 min. intense heat stress (40 °C) and a significantly faster recovery than offspring of control mothers. Offspring matched to their mother's environment had significantly lower but similar corticosterone response to a single ACTH injection and significantly higher but similar respiratory quotient compared to offspring mismatched to their mother's environment. These results demonstrate that some effects of maternal heat stress are not express if offspring are raised in the same stressful environment and that maternal heat stress might prepare offspring for high temperatures in their postnatal environment.

PRONOUNCED MATERNAL EFFECT ON HYBRID GENE EXPRESSION IN ARABIDOPSIS

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Understanding how hybridization influences phenotype is of major importance for evolutionary biology. Theoretically, phenotypic differences are expected to be controlled mainly by gene regulation, which can be environmentally or genetically determined. However, until recently it has been difficult to accurately measure expression levels and determine whether hybrid gene expression are caused by additive (intermediate expression levels between the parents), dominance (expression levels equal to one of the parents) or parental effects (gene expression similar to that of either the mother or the father). Here we use high throughput RNA-sequencing (Illumina) to test the different expression hypotheses and evaluate gene expression in hybrids between two differentiated populations of *Arabidopsis lyrata*. Our results showed (i) broad differential expression between populations (9573 significant genes) and (ii) a very strong maternal effect on hybrid gene expression (94.7% of genes followed the expression of the mother rather than the father plant). In plants, where seed dispersal is limited and the developing seedlings will experience similar environmental conditions as their mothers, such maternal effects are expected to be highly beneficial as they can facilitate local adaptation.

Symposium

15. Attack and Defense: Evolutionary and Ecological Consequences of Individual Variation

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3, 4

Organisers: Anna-Liisa Laine and Pedro Vale

Invited speakers: Meghan Duffy and Marcel Salathé

Description:

Understanding disease spread and host-pathogen co-evolution are central questions in disease biology. Accurate predictions of these processes are made more difficult due to individual variation in defense and attack strategies of hosts and pathogens. This symposium will bring together theoretical and empirical approaches across a broad range of biological systems, to embrace individual level variation in host-pathogen interactions, and its population level consequences.

D20SY15IT10:30R4

SELECTION BY PREDATORS ON WITHIN HOST GROWTH PATTERNS OF TWO NATURAL PARASITES OF DAPHNIA

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Predators can strongly influence the ecology and evolution of host-parasite interactions in a number of ways. Here, we explore the possibility that predators can select on patterns of within host growth by parasites. Are parasites with faster rates of growth within hosts favored in high predation environments? Are there tradeoffs associated with faster proliferation within hosts? We examined how two horizontally transmitted, natural parasites (the bacterium Pasteuria ramosa and the yeast Metschnikowia bicuspidata) of the crustacean Daphnia dentifera varied in their within-host proliferation, equilibrium spore number and transmission rate. We found that both parasites showed logistic growth inside hosts, but that Metschnikowia completed its within host growth much more rapidly than did *Pasteuria*. Bluegill sunfish (*Lepomis macrochirus*), an important natural predator of *D*. dentifera, actively chooses both Pasteuria-infected and Metschnikowia-infected hosts over healthy hosts as prey, and spores can survive and remain infectious after passage through the fish guts. Therefore, predation of infected hosts serves primarily to truncate the time available for a parasite to develop within the host. We selected Pasteuria and Metschnikowia in environments simulating either high or low predation on infected hosts. We found that *Pasteuria* responded to simulated high predation by accelerating within host growth. However, this came at a cost: *Pasteuria* that grew faster yielded fewer mature spores (and these spores were marginally less infectious). Conversely, simulated predation did not affect Metschnikowia infection traits; this result was expected, given that we have previously been unable to find any quantifiable variation in *Metschnikowia*. Overall, we conclude that predation has the potential to mediate the ecology, epidemiology and evolution of disease in a Daphnia-multiparasite system.

D20SY15IT15:45R4

HETEROGENEITY IN CONTACT NETWORKS AND ITS EFFECT ON DISEASE DYNAMICS

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Host contact networks are the roadmaps of infectious disease spread. Much theoretical work has clarified which structural aspects about the networks are important for disease dynamics prediction, but progress has been hampered by a lack of data. Recently, however, novel technologies have allowed to measure host contact networks in great detail. In this talk, I'll give an overview of the field with equal time devoted to theoretical and empirical results.

D20SY15RT11:18R4

VARIATION IN DISEASE EPIDEMICS ACROSS COLONIES

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Colonies of social insects are paradigmatic study subjects for how diseases spread in social groups. Infections are typically acquired naturally from an external source and passed on to the colony members whilst the colony grows and develops through its normal cycle. Here, we report on the spread of an infectious pathogen, the trypanosome Crithidia bombi inside colonies of its host, Bombus terrestris. We show how extensive variation leads to very different outcomes across groups; furthermore, we demonstrate how the parasite population changes genetically as the epidemics unfolds. The data will be embedded in a more general conceptual framework.

D20SY15RT11:42R4

A STUMBLING RED QUEEN: HOST-PARASITE COEVOLUTION HANDICAPPED BY FEEBLE MALES

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Our work highlights the potential influence of intra-specific variations among the sexes in immunocompetence on host-parasite coevolution. In particular, the Red Queen hypothesis proposes that coevolving parasites select for outcrossing in the host. Outcrossing relies on males, which often show lower immune investment as a consequence of sexual selection. Here, we show that such sex-specific variation in immunity significantly interferes with parasite-mediated selection. Two independent coevolution experiments with *Caenorhabditis elegans* and its microparasite *Bacillus thuringiensis* produced a decreased yet stable frequency of outcrossing male hosts. Subsequent tests verified that male *C. elegans* suffered from a direct selective disadvantage. In the presence of its microparasite, males showed lower survival, decreased sexual activity, and altered escape behavior. Each of these responses can reduce outcrossing frequencies. At the same time, males also offered an indirect selective benefit, because male-mediated outcrossing increased offspring resistance. As such intra-specific variations in immunity are widespread among animals, the resulting interference of opposing selective constraints may impose a fundamental limit to host adaptation during antagonistic coevolution.

D20SY15RT14:00R4

DECIPHERING THE IMPACT OF PLASMODIUM PARASITES ON BIRD ODORANT PROFILE: WHAT MAKES INFECTED BIRDS MORE ATTRACTIVE TO MOSQUITOES?

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It was recently demonstrated, using the avian malaria system (Plasmodium relictum SGS1 lineage), that infected birds are more attractive than non-infected birds to host-seeking Culex pipiens mosquitoes (Cornet et al. 2013 Ecology Letters). These results suggest that malaria parasites might manipulate bird traits that are used by mosquitoes to locate their host. This previous experiment eliminated visual or behavioural cues as potential signals, leaving olfactory cues as the most likely mechanism responsible for this enhanced attractiveness. To elucidate the proximal mechanisms of this fascinating manipulation, we characterised bird odorant profiles before and after a Plasmodium infection. The volatile organic compounds (VOC) emitted by hosts were captured on a solid phase (odour trap) using an innovative experimental setup. Gas chromatography was used for identification (mass spectrometry) and quantification (flame ionization detector) of VOCs. Cx. pipiens mosquito feeding preference (between infected and non infected birds) was simultaneously recorded following the protocol described in Cornet et al.. Here we correlate bird odour profiles and mosquito choice measurements, in order to identify VOC candidates susceptible to manipulation by Plasmodium parasites to render their hosts more attractive to mosquitoes.

D20SY15RT14:24R4

IMPLICATION OF WOLBACHIA ON AVIAN MALARIA (PLASMODIUM RELICTUM) TRANSMISSION BY CULEX PIPIENS MOSQUITOES

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In recent years, there has been a shift in the one host one parasite paradigm with the realization that, in the field, most hosts are co-infected with multiple parasites. This question is particularly relevant when the host is a vector of diseases, because multiple infections can have drastic consequences for parasite transmission at both the ecological and evolutionary time scales. *Wolbachia pipientis* is the most common parasitic microorganism in insects and as such it is of special interest for understanding the role of coinfections in the outcome of parasite infections. We have investigated whether *Wolbachia* can modulate the *Plasmodium* infection success and its effect on different life history traits of mosquitoes, such as adult's size, fecundity and on what is, arguably, the most important component of the vectorial capacity of mosquitoes: their longevity. For this purpose, and in contrast to recent studies that have focused on mosquito-*Plasmodium* and/or mosquito-*Wolbachia* combinations not found in nature, we work on a *Wolbachia-Culex pipiens-Plasmodium* triad with a common evolutionary history. To explore different mechanistic explanations for our results, we have also carried out different experiments in which we investigated whether *Wolbachia* or *Plasmodium* can modulate the energetic budget and the immune response of mosquitoes. Further, we discuss different evolutionary explanations as well as their consequences for *Plasmodium* transmission.

D20SY15RT14:48R4

WHAT'S YOUR GUT FEELING? CO-INFECTIONS DRIVE PARASITE EVOLUTION IN C. ELEGANS

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Host-parasite evolutionary dynamics are typically considered as binary interactions. In nature, most organisms are infected by multiple parasites ("co-infection"), and evolution will thus be driven by a network of antagonistic interactions. Given the fitness consequences of within-host competition and host immunity, we hypothesized that co-infections would change the trajectory of parasite evolution. We tested this using experimental evolution of a lab community – worm host and co-infecting gut bacteria, Staphylococcus aureus and Enterococcus faecalis. Co-infection altered the evolutionary trajectories of both parasite species. Additionally, we find that evolving resident gut bacteria may aid in defending the host from virulent co-infecting parasites. We conclude that understanding and predicting host-parasite evolution requires knowledge of other parasites in the system.

D20SY15RT15:12R4

EFFECTS OF PATHOGENS AND FOOD ON HOST POPULATION DYNAMICS IN DAPHNIA MAGNA POPULATIONS

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In nature, hosts are exposed to a multitude of diseases and a wide range of food qualities and quantities. Hereby, hosts and diseases are in a constant co-evolutionary struggle, which might be influenced by the hosts' food intake. This could lead to non-linear responses in host-pathogen interactions. These host food intake dependent host-pathogen interactions on individual level could be translated to the dynamics in populations. Nevertheless, food and disease effects are usually studied independently. So far, prediction models from host individual to population level exist, which either predict food or disease effects, but not effects of both factors at the same time. To fill this gap, a new mathematical model was developed. Due to its characteristics, the model can additionally be used to inversely predict individual level host parameters from population parameters. The given predictions were then compared to the results of life history and population experiments. Therefore, we tested the crustacean model organism *Daphnia magna*, which offers a wide range of pathogens and foods. We concluded, that population parameters can be successfully predicted from single host parameters and *vice versa*, depending on specific host, disease and food properties.

D20SY15RT16:33R4

GENOTYPE AND SPATIAL STRUCTURE SHAPE PATHOGEN DISPERSAL AND DISEASE DYNAMICS AT SMALL SPATIAL SCALES

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Many devastating pathogens are passively dispersed, and their epidemics are characterized by variation that is typically attributed to environmental factors. Here, by combining laboratory inoculations with wind tunnel and field trials using the wind-dispersed pathogen Podosphaera plantaginis, we demonstrate striking genetic variation affecting the unexplored microscale (<2 m) of epidemics. Recipient and source host genotypes, as well as pathogen strain explain 6-33% of variation of the three key dispersal phases: departure, movement and settlement. Moreover, we find genotypic variation affecting group size of the pathogen dispersal unit, ultimately resulting in increased disease development on hosts nearby the infection source. Together, our results show that the genotypic variation may generate considerable variation in the rate of disease spread through space and time with disease hotspots evolving around initial foci. Furthermore, the extent of genetic variation affecting the entire dispersal process confirms that these traits may be targeted by selection.

D20SY15RT16:57R4

PARASITES IN PATCHY ENVIRONMENTS: GENETIC INFLUENCE ON PARASITE DISPERSAL, LOCAL SPREAD AND ESTABLISHMENT

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Because host populations are generally sub-divided, the epidemiology of a parasite often depends on its ability to disperse among host sub-populations and successfully invade them. To investigate the factors that determine host-mediated parasite dispersal and its subsequent transmission we carried our microcosm experiments with the bacterial parasite *Holospora undulata* and its host, the fresh-water ciliate *Paramecium caudatum*. We found that (i) the dispersal of infected hosts largely depended on both host and parasite genotypes (but not their interaction), suggesting disease spatial dynamics varies with the genetic composition of local sub-populations. Besides, (ii) shortly after the arrival of infected hosts into a healthy population parasite success was determined by the genotype of the carrier hosts and its interactions with that of the parasite and the resident hosts. This further highlights the importance of host genotype for parasite dynamics among host sub-populations. (iii) After a couple of transmission cycles, parasite genotype, in combination with resident host genotype, gained influence, matching classical ideas of parasite transmission. It thus appears that different genetic factors play on parasite dispersal between groups of hosts, its short-term spread and longer-term establishment. This study also demonstrates the role of carrier host genotype for parasite dispersal and influences.

D20SY15RT17:45R4

DETERMINING THE EFFECT OF HEPATITIS C GENOTYPE ON VIRUS OUTCOME

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Infection by hepatitis C virus (HCV) leads to one of two outcomes, which varies amongst patients. Either the infection resolves itself within a matter of weeks, or it can persist over several years. It is difficult to ascertain to what extent this outcome is determined by the virus genotype itself using transmission networks, as these tend to be poorly known. Recently, phylogenetic methods have been created to estimate the proportion of set-point viral load that is inherited from one HIV-infected patient to the next. Studies found that up to half the variance in this trait is determined by the virus genotype. Here, we aim to investigate whether we can detect a similar signal in HCV infections. We first simulate inheritance of a binary trait outcome along a given phylogenetic tree to predict how traits gather in groups, and explain how these simulations are used to ascertain the virus effect on the infection outcome. Finally, we apply our method to HCV cohort data from Australia to try and detect an effect of virus genotype on whether hosts will clear the virus rapidly or develop a chronic infection. We also investigate whether key host SNPs, which are known to affect HCV infection outcome, affect this measure of inheritance.

D20SY15RT18:09R4

PHAGE THERAPY MIGHT NOT HELP EVERYONE; BACTERIAL RESISTANCE AND PHAGE INFECTIVITY EVOLUTION CONSIDERABLY VARIES BETWEEN DIFFERENT CYSTIC FIBROSIS PATIENTS

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Phage therapy, the use of viruses to specifically kill only the disease-causing bacteria, could offer an alternative way to fight against antibiotic resistant bacteria. Even though phage selection could lead to increased bacterial resistance, phage therapy has the potential to become an evolutionary sustainable medicine: phages can coevolve more infective and thus retain their efficiency to keep bacteria under control. Here we studied experimentally in vitro the long-term coevolutionary dynamics of model phage therapy by using four natural phage species and *Pseudomonas aeruginosa* bacterial strains isolated from the lungs of ten Cystic Fibrosis patients. We found that bacteria and phages coevolved during the three weeks of cocultivation. However, coevolution was asymmetric by favouring bacterial resistance over phage infectivity, which suggests that phages might not be able to overcome bacterial resistance evolution indefinitely. When ancestral and coevolved phages were applied continuously to ancestral bacterial cultures, coevolved phages were clearly more efficient in reducing bacterial densities with lower corresponding increase in bacterial resistance. These results demonstrate that environmentally derived phages could be used to eradicate clinical bacterial infections, while the effectiveness of phage therapy could be enhanced by coevolving phages more infective in vitro. Despite this potential, our study also shows an important caveat: phages' ability to reduce bacterial densities varied considerably between different patients having very large to negligible effects. As a result, phage therapy is unlikely to work uniformly across all patients and might need to be adjusted case-specifically according to given host and parasite genotypes. Recognising between-patient variation in bacteria-phage coevolution is thus crucial for developing a broad-spectrum phage therapy, which is effective against multiple pathogen genotypes.

D20SY15RT18:33R4

USING MODEL SELECTION TO DEDUCE THE INFLUENCE OF PATHOGEN GROWTH ON THE MORTALITY TRAJECTORY OF THE HOST

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It is well established that individual mortality trajectories are influenced by both innate and external factors. However, we currently have no way of estimating the relative importance of factors that contribute to complex mortality trajectories, such as those which occur during epidemics. Because infection generates heterogeneity between individuals it is difficult to solve even the most elementary epidemiological questions, including whether individuals survive epidemics because they cleared the pathogen or because they never got infected. Here we use an information-theoretic approach to address this problem. First, we constructed a set of demographic models that described plausible influences of infection and cohort heterogeneity on host mortality. Next, we conducted a temperature-specific demographic study of the age-specific mortality rate of roughly 10,000 Drosophila melanogaster topically infected with Metarhizium robertsii, an entomopathogenic fungus. Finally, we estimated the fit of each mathematical model to the mortality data. We found that the all of the best fit and most efficient models in the model set described individual mortality trajectory as being driven by a rapid increase and subsequent decrease in fungal load, which provides substantial evidence that insects do indeed clear fungal infections. We also identified a significant cohort heterogeneity parameter, the estimate of which was large enough to shift the observed mortality peaks earlier and weaker than the underlying infection. This work illustrates the value of model selection in epidemiology and provides a novel method for testing the roles of infection and inflammation in healthy ageing.

D20SY15RT18:57R4

HOST-PARASITE COEVOLUTION: LOCAL INTERACTIONS CAN LIMIT THE SEVERITY OF FITNESS COSTS ASSOCIATED WITH RANGE EXPANSION

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Many host-parasite systems are known to undergo coevolutionary arms races, where reciprocal increases in attack and defence mechanisms can lead to the emergence of broader resistance and infectivity ranges through time. Recent studies have identified that the extent of population mixing can play a crucial role in host-parasite interactions and can shape the coevolution of many traits, including range expansion. Much of this existing theory is based on the analysis of metapopulations, which incorporate a certain degree of spatial structure, but do not capture local interactions between individuals within subpopulations. These local interactions are known to be critical in many epidemiological scenarios, but their role in the coevolution of resistance and infectivity ranges is unclear. Here, we explore how the impact of fitness costs associated with range expansion is affected by the degree of population mixing at these very fine scales. We present results from an individual based model of microbial communities that incorporates a well-established framework of genetic specificity matching empirical observations of range expansion among bacteria and phage. We show that global competition in well-mixed populations leads to rapid selective sweeps, preventing range expansion at high fitness costs. In spatially-structured environments however, we find that local competition and spatial clustering can maintain coevolutionary arms races even when fitness costs are high. These findings highlight the importance of local interactions between individuals in shaping coevolutionary dynamics.

POSTERS

D20SY15PS0111

IS THERE HORIZONTAL IMMUNE TRANSFER IN NON-SOCIAL BUT GROUP-LIVING INSETCS?

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Social immunity has evolved as an additional level of defense in social insects. Since they live in densely populated colonies of related individuals their risk of infection is high. Social immunity therefore is an upgrade in protection against parasitic attack, on top of individual immunity. Of special interest is the horizontal transfer of immunological protection between individuals as a relevant component of social immunity. Since group-living insects, especially when aggregated in high numbers, share similar risks of infection as social insects, we hypothesize, that horizontal transfer of immunity may also be found in group-living, but non-social insects. To test this, we used the Red Flour Beetle Tribolium castaneum. The beetles live in highly populated densities and have been shown to alter their environment by secreting quinones into the flour as a form of cooperative defense against microbes. Moreover, vertical transfer of immunity (i.e. from parents to offspring) has previously been demonstrated in this species. We cohabitated naive beetles with conspecifics that were either wounded (to activate their immune system) or left naïve. By comparing several immune traits, we found that an activated immune status was transferred to conspecifics. The transfer of an activated immune status among individuals might be an important component of the defense level of a population.

D20SY15PS0145

WHICH PROCESSES SHAPE GRAPEVINE MOTH IMMUNE RESPONSE AGAINST PARASITISM?

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Identification of the selective forces shaping immune traits of organisms in natural conditions is a central question in the field of ecological immunology. The immune response is expected to be the evolutionary response to selective pressure from parasites and pathogens. While laboratory studies support this view, few have investigated variation in immune defence in natural populations and identified the putative causal factors of such variation. Here, I will present data showing significant geographical co-variation between levels of innate immune defences of the phytophagous moth, Lobesia botrana, and their infection prevalence by parasitoids among natural populations. I examine the potential causes of co-variation by testing two non-exclusive hypotheses. The first hypothesis is that such covariation between immune defences and prevalence of infection by parasitoids results from a plastic enhancement of immune defences in L. botrana in response to cues indicating the abundance of parasitoids in the environment. As immune defence is costly to maintain and use, the immune system may unsurprisingly exhibit plastic responses to perceived environmental threats, in the same way that phenotypic defences respond to environmental challenges. The second hypothesis proposes that the covariation between immune defences and prevalence of infection by parasitoids corresponds to local optimal investments to the immune system driven by costs arising from the allocation of resources to immune functions and the benefits arising from higher defence to parasitoid attacks. Our results appear to refute the first hypothesis in which moth larvae would be able to adjust their immunity in response to the presence of parasitoids in the environment, but support the second hypothesis in which levels of investment to immune functions is shaped by the costs of immunity and the benefits of immunocompetence against parasitoids.

D20SY15PS0179

RELATING THE OUTCOME OF HEPATITIS C VIRUS (HCV) INFECTION WITH DIFFERENT HOST SNP POLYMORPHISMS IN A HCV-HUMAN INMUNODEFICIENCY-1 VIRUS (HIV) COINFECTED MAJORCAN POPULATION TREATED WITH PEGYLATED INTERFERON PLUS RIBAVIRIN (PEGIFN+RBV).

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HCV infection is one of the main causes of chronic hepatitis, cirrhosis and hepatocellular carcinoma. The HCV-related disease is accelerated in HIV-coinfected individuals. In these patients, the response to antiviral treatment depends on viral and host factors. Among host factors, several genetic polymorphisms (SNPs) have been associated with treatment response. In the present study, we correlated different host SNPs in the IL28b, CTLA4, LDLr, and HFE genes together with mtDNA haplogroups with the outcome of HCV infection and the response to pegIFN-RBV treatment in a Majorcan cohort with HCV-HIV coinfection. We selected sixty-three HCV-HIV coinfected patients and 59 anonymous non-infected controls. Whereas the population frequency of *IL28b* alleles was similar to that found in other European-American cohort, the frequency of the *rs12979860* C allele was lower compared to other Spanish cohorts. The frequencies of *CTLA4* and *LDLr* polymorphisms were similar to those found in other populations. Differences between patient and control cohorts were found for the H63D mutation of the HFE gene. There were no other differences between patients and controls, regarding the frequencies of other polymorphisms or mitochondrial haplogroups. In the HCV/HIV cohort, the most significant data was found in pegIFN-RBV treatment at 4 weeks, where the IL28b rs12979860 (CC) genotype was significantly associated with rapid virologic response, whereas spontaneous clearance was higher in patients with the CTLA4 +49G allele. No significant relationship was found between SNPs in the LDLr, HFE, nor mtDNA haplogroups with response to treatment. In summary, our results highlight the importance of the genetic background of the host on response to therapy in this special cohort.

D20SY15PS0191

GENOTYPIC AND PHENOTYPIC VARIATION IN TRANSMISSION TRAITS OF A TREMATODE PARASITE

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Understanding the processes that maintain genetic variation in transmission traits of parasites is important as they are directly related to evolution of transmission efficiency and virulence. In addition to variation among parasite genotypes, environmental factors such as the condition of the previous host may also affect infection success of the parasite in the next host. The life cycle of many macroparasites includes multiple consecutive hosts and clonal stages that replicate within intermediate hosts. Parasites that have a clonal reproduction stage in their life cycle provide interesting opportunities to test to what extent variation in life-history traits has a genetic basis. This is because both environmental and genetic sources of variation are present in life-history traits among individual clones whereas variation in lifehistory traits of one clone should result only from environmental effects. In this study we measured transmission traits (i.e. production, activity, infection success and survival of the transmission stages) of parasite clones (*Diplostomum pseudospathaceum*), and tested experimentally whether manipulation of the external environment (food deprivation of the snail host) could explain variability in these traits. All measured traits were highly variable among the parasite clones prior to the food-treatment indicating significant genetic variation in these traits. Importantly, the traits were also phenotypically variable as there was marked temporal variation in the traits within each individual clone. However, food deprivation of the snail hosts had little effect on the parasite traits suggesting that a short-term reduction in host resources was not limiting the production rate or performance of the parasite clones . Overall, these results suggest significant interclonal and phenotypic variation in parasite transmission traits that are not affected by short changes in host nutritional status.

D20SY15PS0252

INDIVIDUAL VARIATION IN VIRAL LIFE HISTORY

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Viruses attacking a host face life-history trade-offs in terms of their rate of reproduction (virulence) and their mutation rate, which can lead to advantageous and deleterious mutations that aid in immune escape and transmission or kill the virus. Though it is known that life history trade-offs can have strong effects on mutation and reproductive rates, few models addressing this problem have studied the role of individual variation in viral life history traits. In particular, we investigate optimal reproductive and mutation rates for viruses over the span of their host-pathogen interaction and ask whether individual variation in life history traits can be maintained. To address these questions, we extend population-level research on viral mutation rates and quasispecies interactions, and present the results of mathematical modelling and evolutionary simulations; we conclude by discussing the implications of our results for the study of viral evolution and medical treatment.

D20SY15PS0271

CAN PARASITOID RECOGNITION OF HOSTS BE IMPROVED BY CONDITIONING TO HOST MATE-RECOGNITION CUES?

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Species-specific semiochemicals, such as mate recognition pheromones, are often exploited by predators and parasitoids as cues for recognizing their prey. Generalist predators, unlike their prey, respond to pheromones of many prey species. Therefore, generalist natural enemies used for pest management tend to search for alternative prey. This reduces biocontrol efficiency and harms nontarget species. We tested whether generalist parasitoids increase parasitism on a specific host after conditioning to its synthetic sex pheromone. Parasitoids learn to associate host presence with related environmental cues. We hypothesized that the efficiency of host search and parasitism would increase at repeated encounter with the learned pheromone odor. Two-phase laboratory experiments tested whether the generalist egg parasitoid Trichogramma cacoeciae (Hymenoptera: Trichogrammatidae) can be conditioned to the synthetic sex pheromone of its host, Lobesia botrana (Lepidoptera: Tortricidae). In the conditioning phase, parasitoids oviposited in *L. botrana* eggs in the presence or absence of the host's pheromone. In the test phase, wasps of both treatments encountered additional *L. botrana* eggs, in the presence or absence of the pheromone. Parasitism rates were compared among treatments. Wasps exposed to the pheromone did not increase parasitism rates, neither in the conditioning nor in the test phase. Possibly, the wasps innately responded to the pheromone, which masked their response to conditioning. To examine this possibility, we tried to condition the wasps to rum extract, a neutral olfactory stimulus, foreign to their natural environment. No conditioning occurred in this experiment either, suggesting poor odor learning in *T. cacoeciae*. We now ask whether chemicals, associated with hosts or host plants, attract the wasps when no conditioning occurs. Olfactometer bioassays are underway to identify environmental cues that guide *T. cacoeciae* to its *L. botrana* hosts.

D20SY15PS0278

NUTRIENT AND BACTERIAL DOSES AFFECT VIRULENCE AND HOST-SPECIFICITY OF THE OPPORTUNISTIC FISH PATHOGEN FLAVOBACTERIUM COLUMNARE

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Flavobacterium columnare is a gram-negative bacterial pathogen that causes columnaris disease in freshwater aquaculture. Columnaris outbreaks occur at fish farms during summer months and may cause mortality up to 100 %. Virulence of environmental isolates of *F. columnare* has been found to be lower than those isolated during disease outbreaks at fish farms. In order to understand factors selecting for the higher virulence at fish farms, we studied if the bacterial dose, exposure time (transient or continuous), or nutrients have an effect on the virulence of *F. columnare*. Three *F. columnare* strains were used in two separate experiments: a non-virulent strain B398 isolated from the lake and two virulent strains from disease outbreaks (B185 and B67). In the first experiment zebrafish (Danio rerio) and rainbow trout (Oncorhynchus mykiss) fingerlings were individually infected with bath immersion (transient challenge) with 9 different doses of bacterial strains B185, B398, and a mixture of these strains. In the second experiment the bacteria (strains B185, B398 and B67) were added in three doses directly into aquaria (continuous challenge) where zebrafish and rainbow trout were maintained. Longevity of fish was monitored for five days in both experiments, and the infection verified by bacterial culture from gills.
We found bacterial dose to have a positive effect on mortality of both fish species. Increase in nutrients had a significantly positive effect on columnaris infection and fish mortality. The non-virulent strain was able to infect the fish when introduced in continuous exposure, but not in transient challenge. Our results suggest that the continuous exposure to bacteria at fish farms combined with a high nutrient level can promote virulence also in environmental non-virulent bacteria. In addition, the zebrafish can be used as a functional model host to study F. columnare virulence and infection dynamics in the laboratory.

D20SY15PS0284

LONGITUDINAL STUDY OF INTESTINAL NEMATODE PARASITE DYNAMICS IN FREE-RANGING BROWN MOUSE LEMURS (*MICROCEBUS RUFUS*)

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Longitudinal studies on the parasite community dynamics in free-ranging mammals are challenging. By mark-recapturing 381 individual mouse lemurs over several years in Ranomafana National Park, Madagascar, we have tracked changes in the parasitic nematode community longitudinally. We have collected detailed data on host morphometrics, age, reproductive status, parasites loads, habitat, population structure, and fecal nematodes. Nematoda is the most specious group of intestinal helminths, but their identification is difficult based on morphology alone. Therefore, we have developed a new pipeline to recognize operational taxonomic units (OTUs) by isolating the nematode larvae from feces, pyrosequencing 450 bp segment of 18S and assigning the resulting sequences to OTUs and tentative species. The same protocol was used for collecting parasite data on other sympatric mammals, including lemurs, endemic rodents and invasive species, for example black rat (Rattus rattus). Mouse lemurs hibernate during the austral winter and in post-hibernation individuals the nematode prevalence is markedly lower and the composition differs from the rainy season. During onset of the rainy season, the nematode prevalence was found to reach 95 percent. Our preliminary genomic data shows that mouse lemurs have multiple different OTUs, one of which is the most prevalent. There are substantial differences in the nematode composition in the sympatric species and the endemic rodents have much lower prevalence of the nematodes compared to the mouse lemurs or the invasive species.

D20SY15PS0288

WITHIN-SPECIES GENETIC VARIATION FOR RESISTANCE IN A P-LIMITED ENVIRONMENT: THE DAPHNIA-PARASITE SYSTEM AS A CASE-STUDY

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These last decades, there has been a strong plea to merge biochemical and ecological insights into a field called ecological stoichiometry. Here, ecological interactions are depicted as a flow of essential elements from one level to another. One established principle is that Daphnia suffer reduced growth and survival when fed on P-deprived algae. Moreover, Frost et al. 2008 demonstrated that parasites can intensify this negative effect by increasing their virulence in a P-limited host. As this study considered only one *Daphnia* clone, it remains to be seen how this environment – host – parasite interaction can be generalized to multiple genotypes. In this laboratory experiment, we selected two sets of six Daphnia magna clones and experimentally manipulated (i) food P-availability and (ii) presence of the microparasite White Bacterial Disease (WBD). Both clonal sets were originally hatched from the same sediment core, differing in their depth of isolation. As the selected depth range corresponds to a time span of about 40 years, we expected these sets to differ genetically as a result of historical adaptation towards differences in P-availability. Our results show a significant three-way GH x GP x E interaction. An increasing N:P ratio positively correlated with *Daphnia* mortality. For old clones, the negative impact of WBD was independent of food quality. On the contrary, recent clones suffered more under reduced P-availability when parasites were present. We conclude that the effect of parasites on Daphnia depends on the level of P-limitation and the identity of the considered population.

D20SY15PS0290

ADAPTATION OF A FUNGAL PATHOGEN TO INDIVIDUAL VERSUS SOCIAL IMMUNITY IN ANTS

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Social insects fight diseases not only by the hygiene behaviour and physiological immune system of the individual group members, but also by their collectively performed, social defences ranging from sanitary behaviours, use of antimicrobials and organisational adaptations. Parasites infecting social insect colonies thus have to overcome both individual and social immune responses, which likely exert very different selection pressures on the parasite. To disentangle these effects and to understand the complexity of host-parasite-coevolution in social insect hosts, we performed a selection experiment on parasite adaptation towards solitary reared ants and ants in a social group, i.e. individual immune defence only or both, individual and social defences. We used the Argentine ant, Linepithema humile, and its fungal pathogen Metarhizium as our study system. Metarhizium is a soil-born entomopathogen and obligate killer of a broad range of insect hosts. It was recently found that the previously named M. anisopliae in fact is a species complex comprising a diversity of different Metarhizium species, which differ in ecological dominance and virulence against insects. To reflect this natural diversity, we used a mix of both M. roberstii and M. brunneum to perform ten passages through both solitary and grouped ants (10 replicates each). We found that fungal species composition and virulence changed over the ten generations, revealing a difference in the adaptation potential of the two sympatric fungal species, as well as an effect of the evolution regime. We can thus conclude that the ecological diversity of the different generalist Metarhizium species may reflect adaptations to a diversity of host species, and that the additional group level defences in insect societies adds new selection pressures for their coevolving pathogens with measurable effects on disease dynamics.

D20SY15PS0324

PHENOTYPIC CHANGES IN OAK SEEDLINGS INFECTED BY POWDERY MILDEW: HOST TOLERANCE RESPONSE OR HOST MANIPULATION BY THE PARASITE?

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Plant parasites negatively affect their host fitness by deriving their nutrition from their host. Infection also often affects the physiology of the host leading to changes in morphology, timing of development or fecundity. Plant pathologists have paid much less attention to these life-history changes which could be seen either as tolerance response of the host or as manipulation by the parasite to enhance its transmission, than to resistance mechanisms preventing infection, thus yield loss. We studied the phenotypic changes induced by the oak powdery mildew on its host, Quercus robur in order to assess the effect of infection on host growth pattern. Fifteen half-sib families of oak (2595 individuals) were exposed to three different levels of powdery mildew: low (with fungicides), medium (natural infection) and high (natural infection supplemented by inoculations). Fine monitoring of host growth, phenology and leaf infection were performed over three years. As expected, increasing infection resulted in increasing growth losses in seedlings. More surprisingly, it also had a dramatic effect on seedling phenotypes by causing an accelerated shoot flushing both between and within years. This increased polycyclism provided benefits for both host and parasite: partial compensation of growth losses and extension of the availability of receptive tissues along the season, respectively. Nevertheless, increasing the number of flushes in a season may expose seedlings to higher damage by winter frost. Altogether, these observations are suggestive of both host tolerance and parasite manipulation responses in the "complex phenotype" resulting from the interplay of host and parasite genes.

D20SY15PS0335

EXAMINING THE ROLE OF DSCAM (DOWN SYNDROME CELL ADHESION MOLECULE) IN IMMUNE DEFENCE IN TWO INSECT MODELS

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Individual variation in defence can be based on genetic and phenotypic variation in host immune genes. An effective immune response needs to specifically recognise and act against numerous parasites. One way that a host can evolve diversity in immune recognition and effector molecules is through alternative splicing, potentially resulting in variation in gene expression at the individual level. It has been proposed for crustaceans and insects that the alternatively spliced gene Dscam, might be involved in specific immune responses against diverse parasites. Factors in the immune signalling pathways, Imd and Toll, can affect Dscam splice variants, but evidence from the literature appears to be scarce and inconsistent with respect to the effect of parasites on total Dscam mRNA expression. In a first experiment we therefore tested whether Dscam expression is increased after bacterial immune challenge in the red flour beetle, Tribolium castaneum, and the fly, Drosophila melanogaster. In a second experiment we tested whether there is a life-history cost at the individual level of Dscam knockdown in T. castaneum. We predicted that if Dscam is involved in immunity that it would result in reduced survival after a bacterial immune challenge, and given that Dscam has previously been found to be expressed in the gonads and is vital for nervous system development we predicted that there may be a fecundity cost to the knock-down. Our results take a step towards understanding a little more about the role of this intriguing gene in relation to host-pathogen interactions.

D20SY15PS0351

STRAIN-SPECIFIC IMMUNE PRIMING IN WOOD TIGER MOTH (PARASEMIA PLANTAGINIS)

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Recent studies on invertebrate immunity show that insects possess mechanisms to protect themselves from a secondary infection of a pathogen they have previously encountered. This suggests that invertebrates are able to differentiate between pathogens and mount more specific defence responses than previously thought. However, mechanisms for this specificity remain largely unknown. Here, we report a strain-specific immune priming response in the lepidopteran *Parasemia plantaginis* with enthomopathogenic *Serratia marcescens* bacteria. A major proportion of the moth larvae survived a septic injury with *S. marcescens* strain to which they had been previously orally exposed. Priming with another strain of *S. marcescens* and a similarly gram-negative, but harmless, *Escherichia coli* both failed to provide the protection later in life. We also show that production of caseinolytic proteases is likely a very important virulence factor in orally induced infection in this system. The findings have implications on the evolution of immune system in relatively short-lived insects, as well as on the selective processes entomopathogens may encounter in the arms race with their hosts.

D20SY15PS0463

THE MAINTENANCE OF INTERSPECIFIC HYBRIDIZATION BY PARASITISM IN A FRESHWATER SNAIL

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There is growing appreciation among biologists that interspecific hybridization presents an additional level in which animal evolution can create variability. Yet the long-term persistence of hybrids remains enigmatic, because they have often been labeled evolutionary dead-ends due to their lower fertility and viability. It has been suggested that parasite-mediated selection has the potential to promote and maintain hybridization, by differentially impacting the fitness of host parental taxa and their hybrids. We tested this hypothesis in the *Melanopsis* hybrid complex, the most abundant freshwater snails in Israel that are commonly found to be infected by a variety of trematode species. We sampled 49 independent sites, and recorded snail shell size and gender, snail population density and infection prevalence. We also measured several chemical and physical parameters such as water depth, substrate type, water temperature, salinity, conductivity, pH and dissolved oxygen. We found that hybrids were significantly less infected than their parental taxa. Moreover, we did not find differences between hybrids and parental taxa in any of the abovementioned biotic and abiotic parameters. These results suggest that hybridization in *Melanopsis* may be maintained by parasitism.

D20SY15PS0555

PARASITE-MEDIATED SELECTION PROMOTES CLONAL DIVERSITY IN A FRESHWATER SNAIL

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Sexual reproduction is ubiquitous among multicellular eukaryotes despite the many disadvantages of sexual reproduction compared to asexual reproduction. One of the most prominent hypotheses to explain the ubiquity of sex is the Red Queen hypothesis, which postulates that parasites track common host genotypes, giving rare host genotypes an advantage. Sexual reproduction is advantageous because it results in the production of genetically variable progeny, some of which may have rare genotypes and thus evade infection. However, in the face of genetically diverse asexual lineages, this advantage of sexual reproduction may be eroded and instead sexual populations would be replaced by diverse assemblages of clonal lineages. We investigated whether parasite-mediated selection promotes clonal diversity in 22 natural populations of the freshwater snail Melanoides tuberculata. We found that infection prevalence explains the observed variation in the clonal diversity of *M. tuberculata* populations, while no such relationship was found between infection prevalence and male frequency. Clonal diversity and male frequency were independent of snail population density. Incorporating ecological factors such as presence/absence of fish, habitat geography and habitat type did not improve the predictive power of regression models. Over 10% of the clonal snail genotypes were shared among 2-4 populations, creating a web of 17 interconnected populations. Taken together, our study suggests that parasite-mediated selection coupled with host dispersal ecology promotes clonal diversity. This, in return, may reduce the advantage of sexual reproduction in *M. tuberculata* populations.

D20SY15PS0560

INDIVIDUAL VARIATION IN COLLECTIVE DISEASE DEFENSES OF ANTS

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Pathogen transmission tends to increase with host density, relatedness and interaction frequencies among potential hosts. Disease defenses are thus particularly important in taxa like the eusocial ants, bees, wasps, and termites, which in turn have evolved sophisticated collective disease defenses. These defenses require the collective performance of several individuals. Variation among individuals in the propensity or frequency to perform these tasks, means that colonies may differ in their effective collective disease defenses based on group composition. It is known that honeybee workers respond differently to diseased brood based on their olfactory sensitivity, which has a genetic underpinning (e.g. patriline). A naturally high degree of multiple mating by honeybee queens thus translates into colony-level effects, where colonies that comprise large proportions of "hygienic" workers are more resistant to parasites and pathogens. We could recently show that – also in ants – collective antiseptic behaviors seem to be affected by the underlying genetics, as the response onset to diseased brood differed between genetically homogenous single-queen colonies and diverse multi-queen colonies. To understand whether individual variation in pathogen detection abilities underlies such behavioral responses at the colony level, we tested the olfactory sensitivity of, and variation among, ant workers from genetically homogenous versus diverse colonies.

D20SY15PS0587

LOCAL CO-ADAPTATION BETWEEN A TOXIC PRIMARY PRODUCER AND A HERBIVORE IN THE AQUATIC: *MICROCYSTIS* AND ITS GRAZER *DAPHNIA MAGNA*

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Recently, strong genotype x genotype interactions between the freshwater cyanobacterium *Microcystis* and its grazer, the large zooplankton species Daphnia, were demonstrated, reflecting a strong association: Microcystis may intoxicate and kill Daphnia, but Daphnia populations can graze on *Microcystis* nonetheless, genetically adapting and withstanding its toxins. As this genetic specificity of grazer-prey compatibility is a strong indication of close coevolution between *Daphnia* and *Microcystis*, we hypothesize there is local co-adaptation in this system: in order to persist, Daphnia clones would have to possess the right combination of tolerance mechanisms to tackle the exact set of anti-grazer defenses the local Microcystis strains have lined up. In this study we set out to quantify the degree of local co-adaptation between *Daphnia* and *Microcystis* populations on a subcontinental scale (Belgium vs. Sweden). In a small laboratory experiment, designed to cross-compare sympatric and allopatric Daphnia-Microcystis pairs, we exposed two Daphnia magna genotypes from a Swedish and a Belgian lake to two allopatric and two sympatric Microcystis strains isolated from the same lakes as the Daphnia clones in a full factorial design. We found significant patterns consistent with our hypothesis that Daphnia clones are better able to cope with sympatric than allopatric Microcystis strains. From these results, we could conclude that Daphnia magna populations are indeed adapted to the local Microcystis strain composition, and capable of withstanding reasonable amounts of cyanobacteria in their diet.

D20SY15PS0592

AN INVASIVE SPECIES REVERSES THE ROLES IN A HOST-PARASITE RELATIONSHIP

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Host-parasite relationships are often characterized by coevolutionary dynamics of parasite adaptations to exploit their host, and counter-adaptations in the host to avoid the costs imposed by parasitism. Such dynamic can be severely affected by introduction of a non-native species. We used a unique reciprocal host-parasite relationship between European bitterling fish, Rhodeus amarus, and European unionid mussels to study the effect of an invasive mussel species on bitterling oviposition behaviour and its consequences. Bitterling lay their eggs into gills of live mussels and, in turn, mussel larvae parasitize fish. The European bitterling colonized most of Europe relatively recently and parasitize all sympatric European mussels, which are evolutionarily naive and have not evolved strong defences (egg rejection) against bitterling parasitism. The parasitic larvae of European mussels are unable to utilize the bitterling, although readily parasites most other European fishes. The Chinese pond mussel, Anodonta woodiana, recently colonized European freshwaters from the region of high abundance and diversity of Asian bitterling species where it evolved strong adaptations against bitterling parasitism. We found that European bitterling behaviourally responded to A. woodiana as to a potential host, but failed to use it properly for oviposition (one population) or used it but all the eggs were lost due to rejections by the mussel (second population). Another experiment revealed that parasitic larvae of A. woodiana can develop on the European bitterling successfully, and hence effectively reversed the host-parasite relationship between the bitterling and mussels. We also discuss potential long-term consequences on population dynamics of the bitterling fish.

D20SY15PS0601

THERMAL CHANGES ALTER THE OUTCOME OF BEHAVIOURAL INTERACTIONS BETWEEN CO-EVOLVED HOST-PARASITOID SPECIES

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Climate change often triggers the question if species response to a changing environment will enable them to persist. However, species and especially tightly co-evolved ones, do not exist alone and are largely dependent on interactions with others within communities. Here, we use a mechanistic approach to test the general hypothesis that interspecific differences in the response to short-term thermal changes can change the outcome of host-parasitoid behavioural interactions. We measured the effect of temperature (15, 20 and 25°C) on resting metabolic rates (RMR) of the main pest of cereal crops in Western Europe, the grain aphid host pest Sitobion avenae and its main specialist parasitic wasp, Aphidius rhopalosiphi. Also, thermal behavioural responses of host and parasitoid were measured independently and in interaction, since behavioural strategies of both species largely determine parasitism success. At high temperature parasitoids had lower oviposition efficiency and aphids expressed more defensive behaviours. This alteration in behaviour is likely due to the higher RMR of hosts than parasitoids at high temperature, suggesting RMR is a valuable proxy for predicting the direction of change in the outcome of species interactions. We proposed temperature-induced alteration of species interactions to be a mechanism through which climate change affects ecological communities. Since our findings show that relatively modest thermal changes with non-lethal effects can alter interactions in co-evolved species, ecosystem services such as biological control of pest populations, could be drastically affected.

D20SY15PS0612

ECO-EVOLUTIONARY DYNAMICS IN HOST-PARASITE INTERACTIONS

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It is known that rapid evolutionary processes can play a critical role in host-parasite interactions. Growing evidence suggests that rapid evolutionary change can influences species interactions and thus population dynamics. Here we present results from an experiment with freshwater algal-virus systems, where we examine the role of evolution on host-parasite dynamics for dozens of generations. We followed population dynamics in long-term chemostat experiments, where *Chlorella variabilis* populations were infected with Chloroviruses. Additional time shift experiments allowed distinguishing the evolution of new genotypes from phenotypic responses to infections. We found that high virus densities selected for resistant host genotypes but that the resistance comes at a cost of reduced competitive abilities. The trade-off between resistance and reduced competitive abilities resulted then in the coexistence of multiple host genotypes and ultimately in the stabilization of the host-parasite dynamics. Thus rapid evolution of host-resistance not only altered the population dynamics but also the maintenance of different genotypes in the host population. Our results underline the importance of rapid evolutionary change for species interactions and population dynamics even on ecological time scales.

D20SY15PS0635

THE EXPRESSION OF VIRULENCE IN A CRUSTACEAN-BACTERIUM HOST-PARASITE SYSTEM: AGE MATTERS

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Virulence is a major theme in evolutionary biology. Its expression and evolution have widespread implications for the management of infectious diseases, administration of vaccinations and epidemiology. Underlying the theoretical framework used to describe the evolution of virulence is the trade-off hypothesis, which states that increased parasite transmission rate comes at the cost of shorter infectious periods and hence, less time for the parasite to transmit. However, epidemiological feedbacks resulting from host and parasite life history traits that are under selection during coevolution may alter the virulence-transmission trade-off. In the present study we examined the effects of host age on parasite-induced host mortality (virulence), infectivity and proliferation ability (transmission). We used two asexual clones of the freshwater crustacean Daphnia magna as hosts, and two clones of its semelparous endoparasite Pasteuria ramosa. We found that increased host age significantly reduced parasite infectivity, spore production, and the ability of the parasite to castrate its host, though GxG interactions also affected these variables. Nevertheless, the phenotypic expression of virulence did not differ among host age groups. Interestingly, host-parasite combinations that were highly infected in one age group were not necessarily highly infected in other age groups. Regardless of the possible source of these variations (e.g., differences in host defense mechanisms or enhanced immune function in adult Daphnia) our results emphasize the need to incorporate realistic epidemiological conditions when studying the expression and modeling the evolution of virulence.

D20SY15PS0704

SPATIOTEMPORAL RELATIONSHIP BETWEEN MALARIA INFECTION AND ENVIRONMENT

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The recent integration of molecular genetics and geographic information systems has set a new stage for evolutionary studies of host-parasite interactions. It has been shown that spatiotemporal variation, especially in abiotic factors such as microclimate and habitat composition, can have large effects on transmission and prevalence of infection. Understanding the impact of landscape ecology on hostparasite interactions is increasingly important in the light of climate change and habitat fragmentation. However, epidemiological studies still lack information on how biotic and abiotic factors interplay in relation to host-parasite relations. We investigated the spatial distribution of avian malaria (Haemosporidia) infections in two migratory passerines, pied and collared flycatchers (Ficedula spp.) breeding on the Swedish island Öland in the Baltic Sea. We determined the prevalence and identity of infections using molecular tools (nested PCR, sequencing) and subsequently conducted an analysis of the spatial distribution of infection within habitat patches. Digitally derived data on habitat composition and weather conditions were used in our analysis. We report the relationship between malaria prevalence and a suite of abiotic factors, such as distance from habitat edges, water sources, and human settlements. We discuss the potential impact spatial heterogeneity in the distribution of different malaria lineages in relation to environmental factors has on infection dynamics in this system and on host-parasite co-evolution in general.

D20SY15PS0721

SPECTACULAR VARIATION IN IMMUNE RESPONSES OF BUMBLEBEES TO A TRYPANOSOME PARASITE

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Social living offers many benefits, but comes with a greater risk of infection, especially when the groups are genetically homogenous, as in some social insects. Surprisingly, the first social insect genome, the honeybee, revealed a reduced immune system. Honeybees, like many other highly social insects mate multiply, producing a genetically diverse and complex colony for parasites to invade. Bumblebees, however, mate singly thereby producing dense colonies of highly related sisters. The bumblebee Bombus terrestris is commonly infected with a trypanosome gut parasite Crithidia bombi. Different genotypes of bumblebees are susceptible to different clones of parasite, but the factors that produce this complex pattern of host-parasite matching remain elusive. We took a transcriptomic approach to understand how bumblebees respond to this parasite and whether host-parasite matching is driven by gene expression differences. We find exceptionally high variation in gene expression upon infection despite the low genetic diversity among sisters. This variation among individuals may be especially important in producing an immunologically diverse population, buffering the colony from parasite spread. We also find that different host genotypes down-regulate the same genes but upregulate distinct suites of genes upon infection. This pattern suggests a core shared signal of infection, but a unique suite of up-regulated genes. We also find that clones of this parasite alter expression of their hosts differently. Highly infectious clones down regulate important immunological genes such as antimicrobial peptides but colonies differ in the scale of this effect. Host-parasite specificity may be generated by these specific patterns of expression. We discuss how parasites may manipulate host immune responses for their own gain and how variation among workers may contribute to the sustainability of highly related insect societies in a parasite rich world.

D20SY15PS0725

SEASONAL CHANGES IN IMMUNE DEFENSE IN THE ANT FORMICA EXSECTA

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Eusocial insects are favorable targets for parasites due to high relatedness within a colony and a large amount of interacting individuals. Consequently, eusocial insects evolved a strong defense against parasites on an individual, as well as a social level. When resources are scarce, individuals and colonies are weakened and opportunistic pathogens might be able to infect the hosts, which face a trade-off between energy saving and immune defense. Different demands for energy use during the active season and hibernation are likely to influence this trade-off. This possible trade-off might even be stronger when resource availability is low and starvation imposes additional stress on the individual. Here we investigate seasonal differences in regulation of immune defenses under starvation in the ant *Formica exsecta*. We used bioassays coupled with gene expression analysis of immune, stress and storage protein. In order to estimate the organism's capability to fight off infections, depending on seasonal and nutritional status, entomopathogenic bacteria *Serratia marcescens* and *Pseudomonas entomophila* were used.

D20SY15PS0739

PREFERENCE FOR INFECTION: PATHOGEN MANIPULATION OR HOST DECISION?

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Disease dynamics in social groups are affected by selection both at the individual and group level. This is particularly true in social insects, where highly related individuals frequently interact within densely populated colonies in a confined nest environment, therefore increasing the pathogen transmission and the risk of infection. In response to the high selection pressure imposed by pathogens, insect societies have evolved a battery of cooperative social defenses that complement the immune response of individual group members. In such social immunity, avoidance and nest hygienic behaviors have been recognized as playing a major role in reducing pathogen uptake and exposure risks. Here we show how colonies of the highly invasive ant Monomorium pharaonis fail to avoid pathogens during the critical step of nest site selection. Surprisingly, when presented with the choice of a nest containing nestmates overgrown with sporulating mycelium of the entomopathogenic fungus Metarhizium brunneum (infected nest) and a nest containing nestmates killed by freezing (uninfected nest), experimental colonies preferentially moved into the infected nest (83%, P < 0.001). Colonies did not show a preference when presented with the choice of a corpse-free and an infected nest (61%, P = 0.28), but colonies were more attracted by a corpse-free rather than an uninfected nest (84%, P < 0.01). Together, our results suggest a masking-effect produced by the pathogen, possibly increasing its transmission. Alternatively, pathogen contact can lead to socially inoculating nestmates, thereby increasing colonywide immunity. We can therefore not rule out the possibility that colonies actively chose nests with pathogens to increase their survival chances when exposed again later in life. Colony-level selection operating on such trait can have important implication in shaping both host-disease dynamics and population structure.

D20SY15PS0794

MECHANISMS OF CATERPILLAR BEHAVIOURAL MANIPULATION BY TWO VIRUS SPECIES: SPOT THE DIFFERENCE

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Baculoviruses are known to manipulate the behaviour of their caterpillar hosts, by inducing hyperactivity and altered climbing behaviour. To date, however, knowledge on parasitic genes governing such behavioural manipulations is scarce. Previously we have shown that the baculovirus *Autographa californica* nuclear polyhedrovirus (AcMNPV) induces hyperactive behaviour in *Spodoptera exigua* caterpillars, and that the viral protein tyrosine phosphatase (*ptp*) gene is a key player in the induction of hyperactivity. However, this gene is present in only a subset of phylogenetically related baculoviruses, while it is hypothesized that also baculoviruses that do not carry *ptp* are able to manipulate behaviour, possibly induced by a different viral gene. We compared behavioural changes in *Spodoptera exigua* caterpillars induced by two baculovirus species: the generalist AcMNPV (carrying *ptp*) and the specialist SeMNPV (not carrying *ptp*). We discuss the possible (difference in) mechanism that these two viruses use to manipulate behaviour in the context of host-pathogen coevolution.

D20SY15PS0811

A TURANDOT FAMILY GENE PROMOTES IMMUNITY AGAINST SEXUALLY TRANSMITTED FUNGAL INFECTIONS IN DROSOPHILA MELANOGASTER

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Although it is well known that mating increases the risk of infection, we do not know whether females have adaptive responses to mitigate the fitness costs of sexually transmitted infections (STIs). The major problem has been that we lack a suitable model host/pathogen system for studying the costs of STIs. Recently, Turandot-M (TotM), a member of the Turandot family of genes involved in immune and stress response, has been shown to be upregulated in female *Drosophila melanogaster* when they hear male courtship songs. Here we use the Gal4/UAS RNAi gene knockdown system to test whether TotM provides survival and fecundity benefits for females that mate with fungus-infected males. We show that when the fungus, *Metarhizium robertsii*, is sexually transmitted it reduces female reproductive output across all fly lines. By comparing the knockdown line (Gal4/UAS-TotM) with control lines (Gal4/+ and UAS-TotM/+), we found that TotM provides a survival benefit for females infected with an STI and a survival cost for healthy females. Interestingly, TotM does not provide a benefit for females under direct topical infection with *M. robertsii*. Together these results show that TotM plays a previously-overlooked role in defence against STIs and it suggests that females use auditory cues to anticipate the immune challenges that come with mating.

D20SY15PS0881

AVIAN MALARIA PARASITES AND PATERNITY IN THE BLUE TIT

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Monogamous bird species commonly adopt extra-pair mating as an alternative reproductive strategy. Since such behavior is potentially associated with very high costs in terms of desertion by the social male, it should be balanced by benefits, which may be either of direct or indirect origin. In the latter case, females may benefit from engaging in mating outside the pair bond if extra-pair males pass to the offspring genes of superior quality. One of the main determinants of individual quality is resistance to parasites. Therefore, if the social mate of the female is parasitized, she might be willing to seek resistance genes for her offspring through matings with other males. However, despite the strong theoretical prerequisites, robust empirical data verifying this prediction is lacking. Here, we test this hypothesis using infection status with malaria parasites as an index of genetic resistance. Blood parasites causing avian malaria (genus *Plasmodium* and *Haemoproteus*) are widespread among passerine birds and they may negatively affect fitness of the host by either reducing survival or reproductive success. We verified whether the occurrence of extra-pair offspring in the brood was related to infection status with avian malaria parasites of the social father in the wild blue tit (Cyanistes *caeruleus*) population characterized by relatively high frequency of extra-pair matings (40% of nests contain at least 1 extra-pair offspring) and approximately 60% infection rate with malaria parasites among adult males and females. Additionally, we examined whether the occurrence of extra-pair young is affected by infection status of the mother and the interaction between the infection status of both social parents.

D20SY15PS0892

GENETIC SPECIFICITY IN AN INSECT VIRUS INTERACTION

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Coevolutionary arms races are predicted to lead to rapid evolutionary changes in both competing partners, with the host evolving resistance mechanisms giving rise to counter-adaptations in the parasite and vice versa. This process may lead to specificity in genetic interactions, where the outcome of an infection depends on the genotype of both the host and parasite. We have studied the genetic basis of resistance to parasite replication in an insect virus. The sigma virus is a natural parasite of Drosophila melanogaster, which is vertically transmitted and tightly coevolved with its host. We mapped a major-effect polymorphism previously known as the *ref(3)d* locus to a region containing the two paralogous genes *CHKov1* and *CHKov2*. In a panel of inbred fly lines, we found that a transposable element insertion in the protein coding sequence of *CHKov1* is associated with increased resistance to infection. This resistant allele has rapidly increased in frequency under directional selection and is now the commonest form of the gene in natural populations. Using genetic mapping and site-specific recombination, we identified a third genotype with considerably greater resistance that is currently rare in the wild. By screening a panel of wild viral isolates, we found that there is indeed evidence for a viral counter-adaptation to this resistance genotype. Candidate mutations for this phenotype include a non-synonymous mutation in the m-gene, which produces the viral matrix. This currently rare viral variant illustrates how parasites can rapidly overcome host resistance mutations.

D20SY15PS0932

MUTUALISTIC PARASITES

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We are going to analyze the hypothesis that organisms may use their pathogens for their own benefit: to infect other organisms of the same species. Across History, one can find several examples of humans gaining advantage in warfare using microorganisms. For example, in the Middle Ages victims of infections could themselves become weapons: they were simply catapulted into towns. We will discuss whether bacteria can use their temperate viruses as biological weapons. Temperate viruses are able to be kept in a dormant stage inside a bacterial cell, being vertically transmitted to bacterial progeny during bacterial replication. Sometimes (but rarely), a virus present in one of these bacterial cells "awakes" and starts self-replicating, leaving the cell in big numbers. Cells bearing dormant viruses (called lysogens) are not re-infected by the same virus. Therefore, these viruses, one integrated in the bacterial chromosome, may be used as <<replicating bacteriocins>>. With *Escherichia coli* bacterial cells and the lambda virus, we present data showing that this is indeed possible. However, the best conditions for that (enabling massive virus multiplication among sensitive bacterial cells) are also the best conditions for the integration of the virus into the bacterial chromosome (lysogenization). We further discuss the generality of this experimental result.

D20SY15PS1038

OUT RUNNING THE RED QUEEN? HOST EXTINCTION AS A RESULT OF HOST- PARASITE COEVOLUTION

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One prediction of host-parasite coevolution is that the virulence of the pathogen may change as it adapts to its host in a way that maximises its reproductive success and transmission potential. This has indeed been demonstrated in several controlled laboratory coevolution experiments, with both reductions and increases in virulence being seen. The majority of coevolution experiments, however, use discrete host generations taking only a limited number of usually uninfected hosts to start the next generation. Parasite generations are also often, to some extent, discrete, often being extracted from dead or infected hosts to start the next generation, discarding any spores or longer lasting parasite stages that made their way into the environment. For spore forming parasites, however, this is quite different from the likely natural scenario, where spores can survive for long periods of time in the environment and therefore may obtain the greatest evolutionary benefit by getting as many spores into the environment as possible. If under experimental conditions only spores from dead individuals are taken to the next generation, this benefit is not realised, potentially resulting in an outcome, substantially different to what might be seen in nature. I carried out a coevolution experiment using the red flour beetle, Tribolium castaneum, and its natural microsporidian pathogen, Paranosema whitei, with an overlap in host generations. Flour from the beetles' environment, including any spores which made their way into it, was also transferred from generation to generation. Three treatments with different starting pathogen concentrations and a pathogen free control were used. In all cases coevolution resulted in extinction of the host population, with a pronounced increase in virulence being seen.

D20SY15PS1050

EXPLORING NATURAL VARIATION OF ENTOMOPATHOGENIC FUNGI IN CONTROLLING MALARIA MOSQUITOES

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Malaria is a vector-borne disease that causes a huge burden to humanity, causing around 600 thousands deaths each year, of which 80% are children under the age of five years. Action to interrupt malaria transmission can be achieved by drugs and vector control. Current tools for vector control involve the use of insecticides though impregnated bed nets (ITNs) or indoor residual spraying (IRS). Unfortunately, these strategies have limitations because of the rapid spread of insecticide resistance in mosquitoes. New tools have to be employed to effectively combat malaria, such as entomopathogenic fungi that could be more sustainable and safe for the environment. To make this approach more effective and evolution proof, is crucial to study the components and mechanisms of fungal virulence by exploring the potential development of fungal resistance by the mosquito. We focus on the cosmopolitan fungus Beauveria bassiana which has already been successfully tested in field-based trials. In this study we characterized the natural variation of virulence of 20 isolates of *B. bassiana* that come from distinct regions of the world against the malaria mosquito Anopheles gambiae. Our results showed pronounced differences in virulence between isolates. Interestingly, there was no clear trend concerning geographic origin of isolates and virulence. We further evaluated the relationships between virulence and several fungal characteristics such as spore size, hyphal growth, UV resistance and conidiation production, among others. The contribution of these components to the overall virulence is elucidated. This study highlights the need of further multidisciplinary approaches for understanding the interaction among the many mechanisms shaping the virulence of *B. bassiana* against the insect *An*. aambiae.

D20SY15PS1106

ON THE MULTITROPHIC INTERACTIONS BETWEEN A FUNGUS, A PLANT AND TWO INSECT SPECIES

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In the archipelago of Åland, Finland, lives the Glanville fritillary butterfly, Melitaea cinxia– a wellknown model species for metapopulation studies. One of M. cinxia's host plants, the ribwort plantain Plantago lanceolata, is also host to the powdery mildew fungus Podosphaera plantaginis. Both M. cinxia and P. plantaginis persist as metapopulations in Åland and some local butterfly populations inhabit patches where P. lanceolata is infected by P. plantaginis. It is known from previous studies that development of M. cinxia larvae is affected by mildew infected diet and, interestingly, a parasitoid wasp, Cotesia melitaearum, that uses M. cinxia as its host, is also affected by the presence of mildew in the host diet. Thus, the powdery mildew indirectly affects the higher trophic levels in this community of different hosts and exploiters. In an ongoing study, we investigate the ability of butterflies and parasitoids to tolerate the mildew effects by taking the natural variation within local populations into account. With a feeding experiment, we compared larvae from butterfly populations that have, or have not, been exposed to mildew infection in the past years. Preliminary results indicate that larvae groups originating from populations with different mildew history, responded differently to the diet treatment. However, this interaction between diet and population history does not show a clear-cut pattern and might need to be interpreted with caution. Interestingly, if population mildew history is not taken into account, there is an interaction effect by parasitism and diet on weight at diapause. In addition, parasitised butterfly larvae in general develop faster and weigh more at diapause than non-parasitised larvae. Currently, the effects on C. melitaearum itself, of developing in hosts fed on mildew-infected diet, are being addressed.

D20SY15PS1128

UNRAVELING THE GENOMICS OF ADAPTATION OF COLLETOTRICHUM KAHAWAE TO COFFEA ARABICA

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Ecological speciation through host-shift has been proposed in recent years as a major route for the appearance of novel fungal plant pathogens. *Colletotrichum kahawae* is an emergent plant pathogen causing severe epidemics of Coffee Berry Disease on Arabica coffee crops in Africa at high altitude. This specialist pathogen revealed to be an outstanding model of rapid adaptation and ecological speciation due to its strikingly close proximity (< 2000 yrs) to a non-pathogenic and generalist fungal species. The remarkable genetic proximity of *C. kahawae* and those *Colletotrichum* species associated with a qualitatively change in pathogenicity (*C. kahawae* is able to infect green coffee berries while the closely related species are not) creates a unique opportunity to address fundamental questions behind the evolution of pathogenic fungi. This work aims to use a next generation sequencing approach with Restriction-site Associated DNA (RAD) markers, as well as a broad geographic sampling of *C*. kahawae and its closely related non-pathogenic sibling, to perform a SNP-based genomic population study and unveil the genomic base putatively responsible for the rapid adaptation and severe virulence of *C. kahawae*. Revealing, where and how natural selection is acting across the genome could point us into the direction of the underlying genetics that make the patogenicity of C. kahawae unique and specific to green coffee berries. Moreover, understanding the genomic bases behind these specialization processes will allow a more based-informed and effective control not only for *C. kahawae* populations but also for other plant pathogens with the same speciation mechanism.

D20SY15PS1146

GENETICS OF FEEDING BEHAVIOUR IN DAPHNIA MAGNA AND ITS INFLUENCE ON INFECTION RISK

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Pond sediments represent a primary source of infective stages of parasites of planktonic organisms. The time and the type of interaction with the sediment are therefore important determinants of infection risk. The fresh water planktonic crustacean *Daphnia magna* exhibits a highly variable and heritable phototactic behaviour which influences the time spent in proximity of the sediment. It has been shown that clones that spend more time nearer the sediment have a grater risk of infection. Even if suspension feeding is the primary feeding mechanism in *D. magna*, an alternative behaviour has been described in which small amounts of sediment are stirred up and filtered. This behaviour is expected to increase the encounter rate with parasite infective stages harboured in the sediment and, therefore, infection risk. In the present study we are testing this hypothesis by assessing the heritability of the behaviours, exposed to sediment banks of resting spores of the bacterial parasite *Pasteuria ramosa*. The study is being performed on a large number of clones derived from a quantitative trait loci (QTL) panel. This design allows us to explore the genetic mechanisms underlying the feeding behaviour of *D. magna*, an ecologically relevant trait expected to influence infection risk in natural environments.

D20SY15PS1198

USING POPULATION GENOMICS TO UNCOVER THE GENETIC STRUCTURE, ADAPTIVE VARIATION AND EVOLUTION OF HEMILEIA VASTATRIX, THE PLANT PATHOGEN CAUSING COFFEE LEAF RUST

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Coffee leaf rust (CLR) caused by the biotrophic fungus Hemileia vastatrix is the most important disease of Arabica coffee, reaching nowadays a worldwide distribution. Breeding for rust resistance has proven successful, but the highly adaptable nature of the fungus shaped by the dynamic system of hostpathogen co-evolution has been a critical limitation for achieving durable CLR resistance. As a consequence a gradual breakdown of resistance has been observed over the years in many improved varieties in several countries. In the presence of such a serious and constant threat of new evolving pathotypes emerging under a strong selective pressure and becoming epidemically spread on a continental scale, sound knowledge on population genetics can be used to determine the major forces and mechanisms driving evolution of pathotypes, providing ground to understand how host resistance genes are overcome. A project was recently initiated to address these questions using RADseq of *H*. vastatrix isolates comprising a comprehensive along-time coverage of geographical origins and virulence profiles that provides a unique opportunity for detection of temporal variation over the last 50 years. From the data of thousands of SNPs simultaneously genotyped on those samples, we will be searching for phylogeographical patterns and signatures of selection in the genome. This will allow the identification of putative loci/candidate genes of adaptive significance, and provide insights on the mechanisms of pathogen evolution underlying virulence differentiation. In a parallel conventional population analysis of a virulence-related candidate gene, divergent alleles were detected and signs of adaptive relevant genetic variation are being traced, which could lead us on starting to understand rust adaptation. Funded by FCT (project PTDC/AGR-GPL/119943/2010)

D20SY15PS1316

A MICROCOSM APPROACH TO UNDERSTAND THE EFFECTS OF HOST DEMOGRAPHY ON DISEASE TRANSMISSION

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One key aspect of the evolution of virulence is the rate of transmission between susceptible hosts. In theory, a virulent strain may increase in frequency if selection favours its transmission. However, it is not always know what host traits positively select for transmission and how individual variation and population demography affect its evolution. We are working with an experimental system in which specific aspects of infectious disease dynamics can be measured. We use the nematode Caenorhabditis elegans and fluorescent pathogenic bacteria (Salmonella enterica Typhimurium) to tract the spread of infection. In the soil, C. elegans lives in contact with many microorganisms, including pathogenic bacteria. It is known that similar to the mammalian innate immune response, C. elegans is able to mount a response to Salmonella. Despite this, Salmonella can colonise the worm intestine and increases death. Yet, it is not understood if and how the pathogenic effects of Salmonella can contribute the transmission of the disease in a population. We have characterised in detail *C. elegans* demography in response to the pathogen and varying conditions (i.e. age of infection, worm population size and genetic background). As reported before we found that Salmonella reduces worm survival. However, we found no reduction in the reproduction as a consequence of the pathogen, but a strong negative population density-dependence feedback on *C. elegans*. As a consequence of the high bacteria shedding of worms and little effects on its reproduction, Salmonella is very likely to thrive in the population

D20SY15PS1317

ANALYSIS OF ILLUMINA RNA-SEQ DATA FROM A SUSCEPTIBLE VS RESISTANT COFFEE - COLLETOTRICHUM KAHAWAE INTERACTION

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Coffee berry disease (CBD), caused by the fungus *Colletotrichum kahawae*, is considered one of the biggest threats to Arabica coffee production in Africa at high altitude. Some coffee genotypes are known to be resistant to CBD, but the molecular genetic basis of coffee resistance is still unknown. With the purpose of gaining some insights on this process, a RNA Illumina sequencing approach was used to characterize the defense response of two coffee genotypes, respectively resistant and susceptible to C. kahawae, during the early stages of the infection process. Three inoculation timepoints were selected (24, 48 and 72 hpi) and two biological replicates were collected. Twenty four independent cDNA libraries were sequenced and data is being analyzed to assess differential gene expression when comparing inoculated with control samples. The data was trimmed, and two assemblies were made: one with the control libraries and the other with the inoculated libraries. The first was used in expression quantification for both gene and isoform, by using statistical methods. Particular attention was paid to data normalization, using FPKM algorithm in gene quantification, and a Maximum Likelihood statistical model in isoform quantification. The other assembly is being used to discover C. kahawae genes by codon usage frequency (ECLAT). Although fungal transcripts are weakly represented in the samples, we expect to detect new and unknown genes of *C. kahawae*. The identification and characterization of expression differences between these contrasting situations will allow us to understand which genes are potentially involved in the resistance response of coffee to *C*. kahawae. The subsequent identification of the predicted proteins and their location in the metabolic networks could provide new relevant knowledge able to support and improve coffee breeding for resistance to CBD. Funded by FCT (project PTDC/AGR-GPL/112217/2009)

D20SY15PS1319

IMMUNITY EVOLVES WITHOUT EXTRACTING COST FROM OTHER LIFE HISTORY TRAITS IN DROSOPHILA MELANOGASTER

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Maintenance and deployment of immune system are costly in terms of the amount of resource consumed and has been predicted to trade-off with other resource demanding traits, such as reproduction. However, very few empirical studies have investigated this implicated trade-off surrounding evolution of immunity. We were interested in studying the changes in investment patterns of fruit flies when selected for survival (or better resistance) against a different gram – negative pathogen Pseudomonas entomophila. We subjected this long standing idea to test using laboratory experimental evolution approach. In the present study, replicate populations of Drosophila melanogaster were subjected to three selection regimes – I (Infection with Pseudomonas entomophila), S (Sham-infection with MgSO4) and U (Unhandled Control). Flies surviving the infection four days post – infection are the progenitors for next generation. After >5 generations of selection we observed the flies from the I-regime to have evolved increased resistance to P. entomophila in the form of better survivorship upon infection. However, contrary to the theoretical predictions and at least one previous report, we did not find any evidence of trade-offs between immunity and with other life-history related traits, such as fecundity and longevity. Thus, it becomes important to study different host – pathogen systems in order to have a better understanding of evolution of immunity.

D20SY15PS1336

ARE TOLERANT HOSTS MORE LIKELY TO BE SUPER-SHEDDERS OF DISEASE?

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Understanding the causes of individual variation in transmission could improve the success of disease management, by allowing to target the most infectious hosts. Disentangling the causes of this variation is challenging in the context of epidemics, but an alternative is to study disease transmission (or transmission potential) under a variety of genetic and environmental contexts in controlled experimental conditions. During infection, hosts may recover by eliminating pathogens (resistance mechanisms) or by controlling damage caused by infection without eliminating pathogens (tolerance mechanisms). Because they suffer less for a given parasite load, very tolerant hosts may experience infections for longer and have more opportunities for disease transmission. I present experimental data from Daphnia-microparasite and rodent malaria systems testing the link between tolerance and transmission potential. These results may be particularly relevant in understanding the epidemiological consequences of anti-virulence therapies of infection, that essentially increase host health without eliminating parasites.

D20SY15PS1349

FIELD PHYSIOLOGY MEETS GENOMICS: THE EFFECT OF PROLIFERATIVE KIDNEY DISEASE ON HOST PHYSIOLOGY AND THERMAL TOLERANCE IN WILD BROWN TROUT

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Understanding how genetic and environmental factors influence the response of the host against parasites in controlled laboratory conditions has been an active area of research for decades. However, relatively little is known about the physiological effects and genetic mechanisms of the host defence in wild populations in their natural environment. Here, we measured the effect of the proliferative kidney disease-causing parasite *T* . *bryosalmonae* on the physiology and performance of the host, wild brown trout. We estimated the effect of the parasite on host physiology by quantifying kidney swollennes, hematocrit and different white blood cell parameters in 0+ juvenile brown trout and tested whether PKD have negative effect on upper thermal tolerance and aerobic scope of the host. We also constructed a pedigree using microsatellite markers and used animal models to estimate the heritabity of the physiological traits. Finally, we used a high-throughput SNP chip and association mapping approach to identify genomic regions that affect the severity of PKD. This work demonstrates how integration of physiological, quantitative genetic and genomic approaches can reveal novel insights about disease in wild.

D20SY15PS1408

PHAGE RESPONSE TO PROKARYOTIC CRISPR DEFENSE

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Adaptive immune systems in prokaryotes are centered around repetitive loci called CRISPRs (clustered regularly interspaced short palindromic repeat), into which invader DNA fragments are incorporated (reviewed in [1]). CRISPR transcripts are processed into small RNAs that guide CRISPR-associated (Cas) proteins to invading nucleic acids by complementary base pairing [2,3]. Mobile genetic elements that have been incorporated into the CRISPR blacklist can escape host defense through point mutagenesis [4,5]. Escape mutagenesis induces a secondary immune response leading to rapid incorporation of new invader DNA fragments into the CRISPR locus [6]. MGE's can also escape CRISPR-immunity through alternative escape routes, including (partial) deletions of target sequences from their genome [4,5] and expressing CRISPR suppressors [7]. These features lead to a complex and dynamic co-evolutionary arms race. During this presentation, first the insights gained into the molecular mechanism of this intriguing immune system will be briefly discussed [2,3,5,8], followed by a discussion of the role of CRISPR in bacteria-phage co-evolution ([4] and unpublished data). References: 1. Westra ER, et al. (2012) Annu Rev Genet 46: 311-339. 2. Brouns SJJ, et al. (2008) Science 321: 960-964. 3. Jore MM, et al. (2011) Nat Struct Mol Biol 18: 529-536. 4. Semenova E, et al. (2011) Proc Natl Acad Sci U S A 108: 10098-10103. 5. Westra ER, et al. (2012) Mol Cell 46: 595-605. 6. Datsenko KA, et al. (2012) Nat Commun 3: 945. 7. Bondy-Denomy J, et al. (2012) Nature 493:429-32. 8. Westra ER, et al. (2012) RNA Biol 9.

D20SY15PS1430

FITNESS CONSEQUENCES OF INFECTION WITH MALARIA PARASITES IN THE GREAT TIT – THE MEDICATION EXPERIMENT

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Malaria parasites are one of the most common and widespread parasites among bird species. While severe negative effects of these haemosporidians on host fitness have been observed in naïve populations and domestics birds, the effects in wild avian populations have been generally difficult to detect, because the majority of individuals hold chronic infections. To investigate the consequences of infection with malaria parasites for the parameters of reproductive success in the small hole-nesting passerine – the great tit (Parus major), we medicated the group of females at the early stage of the nesting cycle. The study was conducted in the population with a very high prevalence rate with over 80% of females being infected with at least one parasite lineage (genera Haemoproteus and Plasmodium) at the beginning of the breeding season. During two years females were caught at the nest building stage and injected intraperitoneally either with an antimalarial drug - primaguine, or with a physiological salt. Reproductive performance of birds from the two groups was characterized with clutch initiation date, clutch size, egg size, nestling body mass 2 days post-hatching and fledgling body mass and tarsus length. Clutches laid by primaquine- and physiological salt-injected females did not differ in initiation dates, egg number and size nor there was a difference in body size of nestlings. This data suggest the lack of observable fitness effects of malaria parasites in the study population of great tits.

D20SY15PS1437

EVOLUTION OF A GENERALIST ASSEMBLAGE OF BLOOD PARASITES IN A MEGADIVERSE COMMUNITY OF TROPICAL BIRDS

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Studies on host specificity and its underlying mechanisms are fundamental to understand the architecture of biodiversity and to predict the risks of host switching, which are especially important in conservation issues. Here we test the hypothesis that avian *Plasmodium* and *Haemoproteus* blood parasites have a tendency to be generalists when they face a mega diverse host environment, which is expected from the elevated costs of finding a specific host. We investigate if *Haemoproteus* parasites, which are usually more host specific than *Plasmodium* parasites, are more generalist in a megadiverse bird community in Southern Ecuador. We collected a diverse sample set of 345 wild birds, belonging to 89 species and 21 families and detect a total prevalence of 16.2%. We identify 21 parasite lineages with a wide phylogenetic distribution within the lineages known worldwide, of which 17 appear to be new. We report high levels of host generalization for *Plasmodium* and *Haemoproteus*, and the mean host range of *Haemoproteus* in this tropical bird community is the highest documented to date compared to similar samplings throughout temperate and tropical areas. These findings suggest that the evolution of generalist *Haemoproteus* and *Plasmodium* parasites could be favored in megadiverse bird communities. In these environments, generalist parasites may accrue benefits from the amplification effect associated with the exploitation of various host species, while parasite specialization in single host species may be penalized by severely reduced host availability.

D20SY15PS1460

OXIDATIVE STRESS IN BREEDING GREAT TIT (PARUS MAJOR) INFECTED BY PLASMODIUM SPP.

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The physiological mechanisms involved in the cost of reproduction are poorly understood. In addition, the interaction between reproduction and infection with pathogens is still not resolved. Birds infected with pathogen may be more impacted by physiological mechanisms such as oxidative stress, because of the double physiological cost of reproduction and resistance to pathogens. In this case, an increased metabolism is expected and should lead to a physiological stress involving higher oxidative stress driven by an increased in oxidant production, a higher antioxidant recruitment and/or more oxidative damage to biomolecules. Here, we investigated the effect of reproductive investment and infectious status on parental oxidative stress measures in three wild populations of the great tit, Parus major, naturally infected by Plasmodium spp. Different physiological measures involved in both oxidant production and resistance to oxidative stress were taken from parents when chicks were 14 days old: erythrocyte superoxide production, a proxy of erythrocyte mitochondria quantity (cardiolipin content), and erythrocyte membrane resistance to oxidative attacks. We found a sex dependent effect of reproductive effort on membrane resistance. We also found that parental infection with Plasmodium spp. was linked to an increased production of superoxides, which in turn indirectly decreased membrane resistance. This study suggests that breeding birds incur an additional physiological cost of reproduction linked to infection with malaria parasites.

Symposium

16. Evolutionary Biology in China

21 August



Program

Wednesday 21 August

Session(s): 7, 8

Organisers: Roger Butlin and Kai Zeng

Description:

The aim of the symposium is to show-case the expanding field of evolutionary biology in China. The organisers and lead speakers will use it to encourage Chinese scientists to attend the ESEB Congress. This will be an excellent opportunity for the initiation of new, mutuallybeneficial collaborations as well as to raise awareness in both Europe and China of ongoing work and key groups.

D21SY16RT15:45R8

FRAGILITY AND ROBUSTNESS – THE EVOLUTION OF TROPICAL INTERTIDAL COMMUNITIES AS SEA LEVEL RISES AND FALLS

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Sea level changes may strongly impact intertidal communities. In the tropics, their dominant primary producers are the woody plants of mangroves, which are particularly susceptible to such changes. In this study, we surveyed the genetic diversity of 6 mangrove species in the Indo-western Pacific (IWP) region. In all species, the genetic diversity is organized into two regional clades that are separated by the Strait of Malacca. Under repeated freezes (F) and thaws (T) with past glaciations, or the TFT cycle, the two regions experienced phases of isolation and admixture as the Strait opens and closes. The genetic diversity is built up in isolation and shuffled in the admixture phase. Computer simulations of the TFT model show that the historical effective population size in each region is very small, between 200 and 1200. The results support the conjecture of fragile habitats in tropical intertidal zones under rapid sea level changes. Nevertheless, the geographical distributions of haplotypes suggest adaptive complexes being separately built up in the isolation phase, resulting in robust adaptation in the admixture phase and speciation. Hence, the TFT cycle is a double-edged sword that endowed both fragility and robustness on the intertidal communities of IWP. The confluence of several historical factors in the last 5000 years has tilted the balance toward robustness. However, this balance may quickly swing the other way under multiple influences of local human perturbations and global sea level rises.

D21SY16RT16:09R8

PARALLEL PETAL LOSSES WITHIN THE BUTTERCUP FAMILY (RANUNCULACEAE) AND THEIR UNDERLYING MECHANISMS

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Absence of petals, or being apetalous, is usually one of the most important features that characterizes a group of flowering plants at high taxonomic ranks (i.e., family and above). The apetalous condition, however, appears to be the result of parallel or convergent evolution with unknown genetic causes. Here we show that within the buttercup family (Ranunculaceae), apetalous genera in at least seven different lineages were all derived from petalous ancestors, indicative of parallel petal losses. We also show that independent petal losses within this family were strongly associated with decreased or eliminated expression of a single floral organ identity gene, APETALA3-3 (AP3-3), apparently owing to species-specific molecular lesions. In an apetalous mutant of Nigella, insertion of a transposable element into the second intron has led to the silencing of the gene and transformation of petals into sepals. In several naturally occurring apetalous genera, such as Thalictrum, Beesia, and Enemion, the gene has either been lost altogether or disrupted by deletions in coding or regulatory regions. In Clematis, a large genus in which petalous species evolved secondarily from apetalous ones, the gene exhibits hallmarks of a pseudogene. These results suggest that, as a petal identity gene, *AP3-3* has been silenced or down-regulated by different mechanisms in different evolutionary lineages. This also suggests that petal identity did not evolve many times independently across the Ranunculaceae but was lost in numerous instances. The genetic mechanisms underlying the independent petal losses, however, may be complex, with disruption of *AP3-3* being either cause or effect.

16. Evolutionary Biology in China

D21SY16RT16:33R8

MAXIMUM LIKELIHOOD IMPLEMENTATION OF AN ISOLATION-WITH-MIGRATION MODEL WITH THREE SPECIES FOR TESTING SPECIATION WITH GENE FLOW

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We implement an isolation with migration model for three species, with migration occurring between two closely related species while an out-group species is used to provide further information concerning gene trees and model parameters. The model is implemented in the likelihood framework for analyzing multilocus genomic sequence alignments, with one sequence sampled from each of the three species. The prior distribution of gene tree topology and branch lengths at every locus is calculated using a Markov chain characterization of the genealogical process of coalescent and migration, which integrates over the histories of migration events analytically. The likelihood function is calculated by integrating over branch lengths in the gene trees (coalescent times) numerically. We analyze the model to study the gene tree-species tree mismatch probability and the time to the most recent common ancestor at a locus. The model is used to construct a likelihood ratio test (LRT) of speciation with gene flow. We conduct computer simulations to evaluate the LRT and found that the test is in general conservative, with the false positive rate well below the significance level. For the test to have substantial power, hundreds of loci are needed. Application of the test to a humanchimpanzee–gorilla genomic data set suggests gene flow around the time of speciation of the human and the chimpanzee. Key words: coalescent, maximum likelihood, speciation, gene flow, isolation, migration.

D21SY16RT16:57R8

STATISTICAL TEST TO RELIABLY DETECT RECENT POSITIVE SELECTION BY UTILIZING TREE SHAPE AND BRANCH LENGTHS

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Many methods have been proposed in last two decades to search for traces of recent positive selection and the "maximum frequency of derived mutations" (MFDM) test, which is only based on the unbalanced tree topology, is theoretically insensitive to demography, including bottlenecks and expansions. In this study, we analyzed the statistical features of unbalanced tree in the neutral model and further proposed a new neutrality test that integrates the information of tree topology and the branch lengths of unbalanced tree. Simulations based on wide-range parameters demonstrated that the new test is robust to different demographic models and has a high statistical power to detect recent positive selection. Especially, this test does not need the information of outgroup, and thus will not be affected by the misinference of derived and ancestral variants of segregating sites due to multiple hits. This work provides a reliable method that can distinguish selection from demography when singlelocus or genome-wide DNA polymorphism data is available.

16. Evolutionary Biology in China

D21SY16RT17:45R8

FORENSIC PHYLOGEOGRAPHY OF INVASION: SPECIES EXCHANGED BETWEEN THE U.S. AND CHINA

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Biological invasions represent a pressing conservation problem. Population genetics can provide a powerful forensic tool for understanding the geographic distribution of invasive species, including the history of invasion and the likely sites of origin within the native range. With the rise of global commerce and biogeographic similarities, China and the southeastern United States have become a major conduit for exchange of species that have become invasive in their respective introduced habitats. These species provide an ideal laboratory for investigating evolutionary hypotheses on the evolution of invasiveness. We have been studying a number of plant and animal species to provide a robust phylogeographic baseline for the study of invasion: half the species are native to China and invasive in the U.S. and half are native to the southeast and invasive in China.

D21SY16RT18:09R8

NEW MICRORNAS IN DROSOPHILA – BIRTH, DEATH AND IMMORTALITY THROUGH CYCLES OF EVOLUTION

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The evolutionary dynamics of the entire repertoire of microRNAs (miRNAs) has been debated. Disagreements were not resolved partly because new miRNA-like genes emerged constantly but few were evolutionarily or functionally significant. With an extensive search, we identify 12 new adaptive miRNAs that emerged de novo (and often in clusters) in Drosophila melanogaster in the last 4 million years (Myrs). Interestingly, even though they are adaptively evolving at birth, more than 95% of these new miRNAs disappear over time. They serve a useful function, but for only a transient evolutionary period. After 30 Myrs, all surviving miRNAs make the transition from the adaptive phase of rapid evolution to the conservative phase of slow evolution, apparently becoming integrated into the genome. During this transition, the expression shifts from being tissue-specific, predominantly in testes and imaginal discs, to a broader distribution in many other tissues. Unexpectedly, a measurable fraction (20-30%) of these conservatively evolving miRNAs experience "evolutionary reactivation" and begin rapidly evolving again. These reactivated miRNAs start another cycle of adaptive-neutral-conservative evolution that could also lead to death. In conclusion, new adaptive miRNAs go through cycles of evolution, resulting in very slow turnover. Since most new miRNAs evolve adaptively for a transient period of variable length and then disappear, the phenomenon may shed some light on the regulatory roles of miRNAs in general.

D21SY16RT18:33R8

MICRORNA EVOLUTION BY ARMS SWITCHING AND BI-DIRECTIONAL TRANSCRIPTION

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Proper regulation of gene expression and function is in part regulated by non-coding RNAs. One important class of noncoding RNAs, the microRNA, modulates transcript expression activity. To illustrate how microRNAs can contribute to the evolution of animals' biological regulation, here, I will focus on discussing 1) arms switching and 2) bi-directional transcription of microRNAs located inside the Hox complexes (i.e. miR-10 and miR-iab-4/8). Hox genes are essential transcription factors that establish segmental identity on the head-tail axis of all animals. The miR-10 family is highly conserved in sequence and genomic position among the anterior Hox genes of all bilaterians. By investigating the phenomenon where many miRNAs make functional mature products from one or both arms of the hairpin precursor, it is observed that changes in the miR-10 expression and in the dominant precursor arm from which the mature miRNA sequence derives. Since the predicted messenger RNA targets and inferred function of sequences from opposite arms differ significantly, arm switching provides a fundamental mechanism to evolve the function of a miRNA locus and target gene network. On the other hand, current deep sequencing studies have identified more than 9000 animal microRNAs, but only few loci are known to produce miRNAs from both sense and antisense transcripts. In fly Drosophila melanogaster, the iab-4/iab-8 locus encodes bi-directionally transcribed microRNAs that regulate the function of flanking Hox transcription factors. By expression and functional analyses, it is shown that bi-directional transcription of the miR-iab-4/iab-8 is a conserved Hox regulatory mechanism between fly and the beetle Tribolium castaneum. However, there are key differences in the way Hox genes are being targeted. This allows the understanding of how multiple products from sense and antisense microRNAs target common sites.

16. Evolutionary Biology in China

D21SY16RT18:57R8

THE 'OMICS' OF NON-MODEL ANIMALS

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The rapid development of genomic technology has made unprecedented tools available to evolutionary biologists working with non-model organisms. In recent years, my group at BGI-Shenzhen has developed a series of collaborative genomic projects in with European scientists, particularly with the University of Copenhagen. Several of these focus on complex societies of fungus-growing (attine) ants and (macrotermitine) termites. We have recently completed six ca. 100x genomes and overall transcriptomes covering the crown-group-lineages of the attine ants, and we have sequenced the unusually large genome (1.3 GB) of a fungus growing termite and its fungal symbiont. These reference genomes are now allowing functional gene-expression studies and the development of accurate recombination maps and comparative DNA-methylation studies. These will hopefully shed light on some of the major evolutionary transitions (specialization of fungal crop symbionts, increasing caste differentiation, the evolution of multiple queen mating) in the attine ants. I am also co-directing an avian phylogenomic program to resolve a number of long-standing questions. We have completed about 50 new bird genomes and conducted comprehensive analysis to reconstruct phylogenetic history and link genome-wide differences in evolutionary rates and gene family representation to key adaptations that underlie the different ecologies of the major bird lineages.

POSTERS

D21SY16PS0454

THE PHYLOGEOGRAPHY OF A WIDE-RANGING BIRD IN ASIA, THE COMMON PHEASANT PHASIANUS COLCHICUS

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The common pheasant, Phasianus colchicus is distributed throughout temperate regions in East and Central Asia, and had been widely introduced into Europe and North America as a game bird. This species is well adapted to a wide range of environmental and climate conditions. Thirty-one subspecies have been identified based on substantial variation in male morphological ornaments. We investigated the phylogeography and genetic diversity patterns of common pheasant in Asia using mitochondrial and nuclear genes, and male morphological characters. Our analyses revealed five highly divergent evolutionary lineages and the affinity of subspecies to evolutionary lineages corresponds with four predefined morphological groups and a previously undescribed group. The distribution of these lineages reflects geographical breaks of mountains, plains and deserts, and the estimates for the divergence times between these lineages probably predate the last glacial maximum. We further found evidence of extensive genetic introgression between contiguous subspecies within lineages. Taken together, these results suggest that the phylogeographic patterns of common pheasant had been shaped by ancient colonization events and population expansions during postglacial periods. We are currently investigating genome-wide variation related to adaptation and radiation of common pheasant using a RAD-sequencing approach. Furthermore, our study system provides a favorable framework to study the speciation processes under biogeography, local adaptation and sex selection.

D21SY16PS1294

RECURRENT GENE FLOW VERSUS INCOMPLETE LINEAGE SORTING: ALLELE SHARING BETWEEN TWO HYBRIDIZING PINES WITH OVERLAPPING DISTRIBUTIONS

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When closely related species occur with parapatric or sympatric distributions, allele sharing between them might due to retention of ancestral polymorphism because of incomplete lineage sorting and/or recurrent gene flow following secondary contact. One difficulty for studying speciation is to distinguish between the two. Here, genetic polymorphisms at multiple intronic sequences of two closely related pines (Pinus massoniana Lamb. and P. hwangshanensis Hisa) with overlapping distributions were employed to address this issue. First, we compared the patterns of genetic diversity, linkage disequilibrium (LD), differentiation and population structure in parapatric and allopatric populations of the two pines. Approximate Bayesian Computation (ABC) was then employed in defining demographic and speciation models. Parapatric populations harbored higher levels of intraspecific genetic diversity, lower levels of interspecific differentiation and more rapid decay of LD than allopatric populations. Population structure analyses revealed more introgression in parapatric populations than that in allopatric populations. Model selection based on ABC suggested that the two species came into secondary contact about 10000 generations ago and gradually spread to reach the current patterns of overlapping distributions. Taken together, allele sharing between these two closely related parapatric pines might due to secondary introgression rather than incomplete lineage sorting. Symposium

17. Linking Genome Evolution at Different Time Scales

22 and 23 August



Program

Thursday 22 August

Session(s): 9 Friday 23 August Session(s): 10, 11

Organisers: Nicolas Salamin and Marc Robinson-Rechavi

Invited speakers: Chris D. Jiggins and Laurent Excoffier

Description:

The inferrence of evolutionary forces, notably selection, from molecular data, has a long tradition in evolutionary biology. Studies usually focus either on interspecific or on intraspecific data, and thus address disconnected questions. Thanks to advances in genomics and in computational biology, there is increasing interest in bridging the gap between these two fundamental scales of evolution. This symposium will provide exciting discussions and prospects to where next-generation sequencing data can lead us in the understanding of the selective forces acting during evolution. 17. Linking Genome Evolution at Different Time Scales

D22SY17IT10:30R3

GENOME-WIDE PATTERNS OF ADMIXTURE BETWEEN SPECIES DURING AN ADAPTIVE RADIATION

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N/A

D23SY17IT10:30R3

DETECTION OF POLYGENIC SELECTION AT DIFFERENT EVOLUTIONARY LEVELS

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Most approaches aiming at finding genes involved in adaptive events have focused on the detection of outlier loci, which resulted in the discovery of individually 'significant' genes with strong effects. However, a collection of small effect mutations could have a large effect on a given biological pathway that includes many genes, and such a polygenic mode of adaptation has not been systematically investigated in humans or other mammals. We therefore propose to evidence polygenic selection by detecting signals of adaptation at the pathway or gene set level instead of analyzing single independent genes. Using a gene-set enrichment test, we identify genome-wide signals of recent adaptation among human populations as well as more ancient signals of adaptation in the human lineage and in primates.

D22SY17RT11:18R3

SHORT- AND LONG-TERM EVOLUTION OF GENE EXPRESSION AND SEQUENCE DIVERGENCE ON THE AVIAN Z CHROMOSOME

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Sex chromosomes provide unique opportunities to study the fundamental evolutionary forces that act on the entirety of the genome. Because of their unusual pattern of transmission, biological differences between males and females cause sex chromosomes to experience distinct evolutionary environments, which in turn influence coding sequence, gene expression, and even splice forms. The avian Z chromosome is separated into multiple distinct strata, formed by numerous recombination suppression events that have evolved independently across different avian lineages. This provides a unique opportunity to examine the effects of sex-specific selection on the same chromosome across evolutionary time scales. We first mapped out the fine-scale evolutionary history of the Z chromosome across a clade of birds, encompassing roughly 80 million years. Our results show that replicate strata have formed independently across this clade. We then used multiple integrated next-generation datasets to measure gene expression and gene sequence divergence across four species within this clade that experience different sex-specific selection regimes. Our data indicate that male-biased gene expression accumulates over time across Z chromosome strata, with older strata showing greater levels of malebias. This is consistent with the fact that Z-linked genes are more often selected for male-specific effects. We also show that sex chromosome divergence influences sex-specific alternative splicing, leading to the loss of female-specific exons from the Z chromosome.

17. Linking Genome Evolution at Different Time Scales

D22SY17RT11:42R3

GENOME EVOLUTION AND STRUCTURAL VARIATION IN STICKLEBACKS ACROSS DIFFERENT STAGES OF DIVERGENCE

<u>Frédéric JJ Chain</u>¹, Philine GD Feulner¹, Mahesh Panchal¹, Yun Huang¹, Christophe Eizaguirre², Martin Kalbe¹, Tobias L Lenz³, Irene E Samonte¹, Monika Stoll⁴, Erich Bornberg-Bauer⁵, Thorsten BH Reusch², Manfred Milinski¹

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Deciphering the genetic architecture underlying population differentiation and adaptation is crucial for better understanding the process of ecological speciation. To reveal patterns of genome evolution across different stages of population divergence, we characterized genetic variation in three-spined sticklebacks using 66 whole genomes (15x coverage each) from geographically and ecologically distinct populations. This fish species has recently colonized freshwater habitats and undergone substantial and recurrent phenotypic divergence associated with their habitat. We have evaluated the relative importance of several types of genetic variation (SNPs, INDELs, CNVs, inversions and translocations) in the differentiation of genomes across populations and ecotypes. Structural variations cover a larger proportion of the genome than the ~10 million single nucleotide variants. Whereas the majority of variants are shared across several populations, we detect genomic regions of high differentiation between closely related populations. Lineage-specific genes and RNA genes often differ in copy number between individuals and between populations, suggesting a potential role of structural variation such as CNVs in ecological adaptations. We also investigate the relationship between CNVs and different categories of duplicate genes, and evaluate the molecular rates of gene evolution using interspecific data. Taken together, our findings demonstrate extensive genomic differentiation within only a few thousand generations and support a mechanism for the birth and death of new genes via

duplication, highlighting the dynamic nature of genomes. Due to our population sampling design we are able to shed some light on the interplay of ecological and genomic features of populations during adaptive evolution and at different stages of ecological speciation.

D23SY17RT11:18R3

GENOMIC BASIS OF THE ADAPTIVE RADIATION OF *TIMEMA* STICK INSECTS

<u>Moritz Muschick</u>¹, Victor Soria-Carrasco¹, Patrik Nosil¹

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Although most of biodiversity on Earth is thought to be the product of adaptive radiation, very little is known about its genomic basis. Adaptation and speciation with gene-flow, probably common in adaptive radiations, might result in distinctive patterns of genomic divergence between species and populations. But how much do these micro-evolutionary processes determine the observable macroevolutionary patterns of species differences and similarities? Recent field experiments with the stick insect species Timema cristinae from California have demonstrated significant selection on a number of genomic loci when populations were transplanted to non-native hosts or to different elevations, featuring different climates. Building upon these results we here investigate genomic divergence across several *Timema* species and populations, and its correlation with divergent and convergent adaptation. We have sampled over 1500 individuals from 110 populations of 11 species of *Timema* stick insects. This collection comprises samples from 10 different host plants, covering an altitudinal range of 2800 meters. Using restriction site associated DNA tag sequencing and whole genome resequencing we reveal the patterns of genomic divergence between species and populations from similar and divergent environments. We expect to learn if divergence at certain loci is correlated with divergent adaptation, and how this pattern changes with phylogenetic relatedness. Are the demonstrated short-term changes in allele frequency matching the genomic divergence on a macro-evolutionary time scale in Timema stick insects? Answering this question will advance our knowledge about the influence of microevolutionary ecological adaptation on genome evolution in adaptive radiations.

17. Linking Genome Evolution at Different Time Scales

D23SY17RT11:42R3

ANALYSIS OF GENOMIC STRUCTURAL DIVERSITY AND UNCHARACTERIZED NOVEL SEQUENCES IN WOLVES AND DOGS

<u>Javier Quilez</u>¹, Belen Lorente-Galdos¹, Dorina Twigg², Stefan Sirakov², Oscar Ramirez¹, Iñigo Olalde¹, Jonas Berglund³, Carlos D Bustamante⁴, Adam H Freeman⁵, Matthew Webster³, John Novembre⁵, Adam Boyko⁶, Robert Wayne⁵, Carles Vila⁷, Jeffrey M Kidd², Tomas Marques-Bonet¹

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The domestic dog has been widely recognized as an important organism for studying the relationship between selection, genome variation, and phenotypic diversity. Both dogs and wolves have been extensively surveyed using mtDNA, microsatellites, SNPs inferred from genotyping platforms or pooled sequencing or from a handful of individuals with low genomic coverage. Although structural variation, including variation in multicopy gene families, has been recognized as one of the major contributions to differential phenotypes, extensive characterization of this kind of variation in canines is still lacking. We set out to explore the full spectrum of copy-number and structural genome diversity via next-generation sequencing of a diverse panel of high coverage full genome sequencing of 29 canid genomes. The set includes unrelated specimens from all major populations including 14 wolves, 4 coyotes, 10 dog breeds and one basal dog lineage (Basenji). Validations and genotyping by arrayCGH have been performed in a total set of 20 wolves and 7 dogs. Our analyses have found over 170 Mbps of structural variants in the dog and wolves genomes affecting the complete gene structure on ~250 genes. As expected from their demographic histories, the amount of segregating structural variants is lower in dogs than in wolves. We have also identified specific gene expansions and contractions accounting for 150 coding sequences and we can now date the origin of gene duplications. Finally, we also explored the contribution of dog and wolf novel sequences that encompass 4 million assembled sequences that

are not present in the dog genome assembly. The study of those specific expansions will help us to improve our understanding on the biology of these newly expanded gene families and their role in rapid phenotypic changes.

17. Linking Genome Evolution at Different Time Scales

D23SY17RT14:00R3

EQUILIBRIUM AND NON-EQUILIBRIUM DEMOGRAPHIC HISTORY AND THE DISTRIBUTION OF FST: DEVIATIONS FROM THE ISLAND MODEL CAN STRONGLY AFFECT THE CONCLUSIONS OF QST AND FST OUTLIER TESTS

<u>Michael Whitlock</u>¹, Katie Lotterhos¹ ¹Department of Zoology, University of British Columbia, Canada whitlock@zoology.ubc.ca

Local adaptation predicts that selected alleles and traits will differ in frequencies among populations, as each population adapts to its own optimum. Recently, our field has made increasing use of several methods designed to look for loci or traits that have greater divergence among populations than expected by genetic drift alone. Statistical conclusions from QST approaches (in the case of traits) or FST genome scans (for selected loci) depend on the demographic properties assumed by their null models. Typically an island model or a Dirichlet distribution is assumed. We explored through simulation a number of demographic models that differ increasingly from the island model, including several realistic scenarios out of equilibrium. For both QST and FST, the distribution of differentiation measures sometimes is well described by the island model, but often the differences are profound. We show that the differences can cause a great excess of false positives in QST or FST outlier approaches, and we make several suggestions about how to identify and ameliorate these problems in real biological settings.

D23SY17RT14:24R3

POLYMORPHISMS-AWARE PHYLOGENETIC MODELS

<u>Nicola DeMaio</u>¹, Christian Schlötterer¹, Carolin Kosiol¹ ¹*Vetmeduni Vienna, Institut für Populationsgenetik, Austria* nicola.de.maio.85@gmail.com

Comparative analysis of genomes of related species, and of different individuals of the same species, can reveal adaptive trends in the history of the considered taxa, as well as show intensity and genomic variation of evolutionary patterns. However, these intra and interspecific data also bring new challenges, such as the presence of incomplete lineage sorting and ancestral shared polymorphisms. We propose a new POlymorphisms-aware phylogenetic MOdel (PoMo) that relaxes the assumption of instantaneous substitutions of standard phylogenetic approaches. A substitution is hereby modeled through a mutational event followed by a gradual fixation. Our model utilizes both divergence and polymorphism data from different species/populations. By allowing polymorphisms at internal phylogenetic nodes, it also naturally accounts for incomplete lineage sorting and shared ancestral polymorphisms. PoMo can accurately and time-efficiently estimate phylogenetic trees of any shape and dimension, e.g. species trees, population trees, or any combination of those. It can also disentangle the contributions of mutations and fixation biases in substitution patterns. We analyzed synonymous sites in genome-wide alignments of human, chimpanzee, and two orangutan species. Using PoMo, we obtained accurate estimates of mutation rates and GC-biased gene conversion (gBGC) in great apes. We found that both mutation rates and gBGC vary with GC content, determining the well-known differences in substitution rates. Our results are consistent with the presence of directional selection in synonymous sites regarding exonic splicing enhancers. Lastly, we show with simulations that PoMo accurately estimates phylogenetic branch lengths, whereas standard substitutions models present large biases due to ancestral polymorphisms. Furthermore, our methods are more computationally efficient than coalescent-based approaches.

D23SY17RT14:48R3

DETERMINANTS OF PROTEIN EVOLUTIONARY RATES IN LIGHT OF ENCODE FUNCTIONAL GENOMICS

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The aim is to understand how the complex anatomy and developmental processes of animal influence the evolution of protein-coding genes. The influence of different parameters, from gene size to expression levels, on the evolution of proteins has been previously studied in yeast, Drosophila and mammals. Here we investigate these relations further, especially taking in account gene expression and chromatin organization in different organs and different developmental stages. For expression we used a microarray experiment over zebrafish development as well as the RNA-seq data from ENCODE for 22 different tissues of mouse. We also used chromatin accessibility in mouse tissues, and we use ENCODE data to define which transcript is used as reference to compute gene length, intron number, etc. We find strong differences between tissues or developmental stages in impact of expression on evolutionary rate. Over all tissues, an interesting result is that evolutionary rate is better correlated with maximal expression in one tissue then with average expression value over all tissues.

D23SY17RT15:12R3

EVOLUTION OF CORRELATED SITES IN THE MELANOCORTIN SYSTEM

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Ten years ago it has been demonstrated that evolutionarily co-evolving residues in a protein mediate allosteric communication involved in cellular signaling. Recently, it has been observed that co-evolving positions can also explain folding, and key mutations implied in genetic diseases. Although predictions and biological evidence showed that some positions are correlated in the DNA and protein sequences, the evolutionary models used in phylogeny assume that these positions are evolving in an independent fashion. Here we propose a new model that considers co-evolving positions. The model is based on a 16X16 instantaneous rate matrix and three parameters: s, d, w. where s is the rate associated with a transition from a co-evolving combination to a non-co-evolving one, d is the rate of a transition from one non-co-evolving combination to a co-evolving one, w is the rate attributed to a single mutation occurring between two non-co-evolving combinations. To evaluate the new model, we use likelihood ratio test (LRT) between two models: the null model where independent evolution is assumed for each position (i.e. s=d=w) and the dependent model in which co-evolution is assumed. The results show that the null model has a weaker likelihood when two positions are co-evolving, whereas in the case of independent positions, the dependent and the null models have similar likelihoods. In the past decade several methods have been developed to identify co-evolving positions using probabilistic or combinatorial approaches. These models give a score of correlation but they do not distinguish within co-evolving positions, combinations of nucleotides that are indeed co-evolving across the phylogeny. This likelihood-based framework represents a step forward in reconstructing the evolution of coevolving patterns based on a phylogeny with potential applications in evolutionary studies and mutagenesis experiments.

POSTERS

17. Linking Genome Evolution at Different Time Scales

D23SY17PS0219

BGEE, A DATABASE FOR THE STUDY OF GENE EXPRESSION EVOLUTION

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Gene expression patterns (where and when genes are expressed) are a key feature in understanding gene function and evolution. To apply compare results between different model organisms and human, or to study gene expression evolution, a comparative approach must be used, but no tools allow to easily compare gene expression across species. We have thus developed Bgee (Base for Gene Expression Evolution), a database designed to automatically compare expression patterns between animals. This is achieved by i) the aggregation and curation of expression data from different types and sources, to map them to formal representations of anatomies and developments of different species; Bgee release 12 contains curated and quality controlled data for Affymetrix chips EST libraries, and RNA-seq libraries annotated by our curators, as well as in situ hybridizations. ii) the analysis of these data by dedicated statistical tests to define high confidence gene expression patterns. iii) the definition of comparison criteria between anatomies of different species; Bgee curators have designed relationships between more than 5000 species-specific terms, which map to more than 1000 homologous organ groups; the latter are organized in multi-species ontologies (the HOG and vHOG ontologies). Bgee is available at: http://bgee.unil.ch/

D23SY17PS0250

SURPRISING PATTERNS OF ALLELIC DIVERSITY OF V1R GENES IN TOOTHCOMBED PRIMATES

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Vomeronasal receptor genes are of increasing interest to biological fields as disparate as molecular ecology and sensory integration due to their exquisite one-to-one relationships among pheromonal signals, gene expression, and neuronal sensory input. We report the most extensive sample of vomeronasal receptor class 1 (V1R) sequences ever generated for a diverse yet phylogenetically coherent group of mammals, the tooth-combed primates (suborder Strepsirrhini). We have identified more than 1000 unique alleles from across 19 strepsirrhine taxa. Phylogenetic analysis reveals that we have intensively sampled from a single V1R subfamily, apparently unique to the strepsirrhine primates. The subfamily, which we designate as V1Rstrep, retains extensive repertoires of alleles that descend from an ancient gene duplication that occurred prior to the diversification of all lemuriform primates excluding the basal genus Daubentonia (the aye-aye). Comparison of amino acid sequences between the two descendent lineages reveals different amino acid compositions corresponding to the predicted ligand-binding site, and thus potentially, to altered functional profiles between the two. As supported by a three-dimensional folding model, the ligand-binding site is predicted to have different binding capacities in the two clades. The dwarf and mouse lemur species (family Cheirogaleidae) show strong patterns of gene-tree/species tree incongruence whereas V1R sequences in the true lemurs (family Lemuridae) show far greater fidelity to the species tree. This is despite the fact that the true lemur and the dwarf and mouse lemur clades are of similar geological age. These observations imply that different molecular-evolutionary properties are operating within the taxonomic groups, such a differential birth/death rates and/or gene conversion. The majority of V1Rstrep alleles appear to be intact and under strong positive selection, particularly within the transmembrane regions of the gene.

D23SY17PS0310

WITHIN-POPULATION ESTIMATION OF SELECTIVE CONSTRAINT

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Phylogenetic estimation of the ratio of non-synonymous (Ka) to synonymous (Ks) changes is a commonly used method for estimating levels of selective constraint between divergent populations in a protein-coding region. These methods are also frequently used for within-population analysis; however, it is known that weakly deleterious mutations can generate time-dependent behaviour of Ka/Ks. Purifying selection may take many generations to remove weakly deleterious polymorphisms, so that branches near the tip of the tree will be confounded by both long-term evolutionary processes and population genetics. We show, using forward simulation of a Wright-Fisher population, that the standard method of grouping tip branches in a separate partition does not completely correct for this bias. Using comparisons along each branch, we show that a simple filter based on branch-length can correct for the bias. Population genetic theory implies that the filtering condition depends on both population size and selection strength. However, in practice, it depends only on the length of the sequence. These results imply that between- and within-population comparisons can be used without explicit separation.

D23SY17PS0425

EVOLUTIONARY HISTORY OF THE 17Q21 HUMAN POLYMORPHIC INVERSION

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With the aid of novel and powerful molecular biology techniques the study of the structural plasticity of the genome has gained momentum, and one particular subtype of chromosomal rearrangement – inversions – was recently found to be far more common than predicted from classical cytogenetics. Moreover, large inversions have been identified at high frequency in some human populations. One particularly large inversion (900 kb) described in humans and several Great Apes, namely 17q21, has been shown to exhibit 'frozen' haplotypes (H1 and H2) which were originally identified by specific mutations and by their opposite orientations. The fact that they accumulate mutations independently is not necessarily surprising since recombination is expected to be limited between inverted regions. However, three surprising results were also found (i) age estimates of the inverted-associated haplotype (H2) are in the order of magnitude of millions of years, and (ii) the frequency of the inverted haplotypes vary between 5 and 35% in Europe only, (iii) cases were found in which the inversion status was in contradiction with the molecular haplotype, i.e. some H2 haplotypes (as defined by specific mutations) were in the same orientation as H1. These results suggest that (i) the inversion might be recurrent despite its size, or (ii) the polymorphism was kept during a large evolutionary timescale and resisted various speciation processes since it is observed in other Apes. In order to clarify the complex history of 17q21 human polymorphic inversion we are merging NGS data from 14 populations with cytogenetic (e.g. Fluorescent in situ hybridization) and molecular techniques. Furthermore, we will apply population genetics approaches to compare different evolutionary models.

WITHIN-POPULATION VARIATION IN GENOME SIZE IN BRACHIONUS PLICATILIS ROTIFERS

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Genome sizes among eukaryotes are highly variable. While there is consensus that most of this variation is caused by variable amounts of non-coding DNA (e.g. intron length, pseudogenes, transposable elements), controversy still exists about the evolutionary forces driving such differences, in particular about the roles of selection *vs.* drift or mutation. Organisms displaying intrapopulation variation in genome size may offer an attractive system for resolving such a controversy. Here I present evidence for intrapopulation genome size variation in one lineage of the *Brachionus plicatilis* species complex, a small planktonic rotifer. Genome sizes within this population ranged up to 1,2 fold and the distribution followed a unimodal pattern with a mean of 242 Mbp, based on flow-cytometry measurements with *Drosophila melanogaster* as internal standard. Individuals with high and low genome size were genetically highly similar to each other, according to their ITS1 and COI barcoding sequences, and they could be crossed to produce offspring with intermediate genome size. Altogether this suggests heritable within-population variation in genome size. Due to the ease of experimental manipulation and their short generation times, such rotifer populations should allow a "microevolutionary" perspective on genome size evolution.

RELATIONSHIP BETWEEN EVOLUTIONARY RATE VARIATION, PROTEIN SECONDARY STRUCTURE AND CODON USAGE IN PROTEIN-CODING GENES

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The co-translational hypothesis predicts selection for rare codons in loops connecting protein subunits, such as secondary structures [1]. Rare codons are translated at a slower rate, causing translational pauses that may aid in protein folding. Furthermore, synonymous rates vary substantially from site-tosite in many genes and synonymous mutations have been implicated in genetic fitness and several diseases [2]. Despite this, synonymous substitution rates are often assumed neutral. Here we investigate how the synonymous rate is related to secondary structure and codon usage. We studied 912 human and 542 E. coli genes with assigned secondary structures. Orthologs from 14 mammalian and 19 E. coli strains were extracted and site-specific evolutionary rates inferred using models implemented in CodonPhyML [3], an extension of PhyML. Only a weak correlation between codon usage and secondary structure was found, but evolutionary rates correlated stronger with secondary structures, showing that different structures are under different selective constraints for codon choice. Presence of loops with low synonymous rates points to purifying selection on synonymous codon choice, consistent with evolutionary conservation of translational pauses. However, higher average evolutionary rates for loops compared to sheets and helices indicate that, in general, loops are under weaker purifying selection, possibly due to higher lineage heterogeneity. These findings do not rule out the presence of rare codon clusters, but suggest that they are not widespread. [1] Komar, A.A. A pause for thought along the co-translational folding pathway. TiBS, 34(1), 2009 [2] Sauna, Z.E., Kimchi-Sarfaty, C. Understanding the contribution of synonymous mutations to human disease. *Nat Rev Genet*, 2011 [3] Gil, M., Zanetti, M., Zoller, S., and Anisimova, M. CodonPHYML: Maximum likelihood phylogenetic inference under codon substitution models. *MBE*, 2013 (accepted with minor revision)

GENOME-WIDE PATTERNS OF GENETIC VARIATION IN EUROPEAN ASPEN (POPULUS TREMULA)

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The recent advancement of next-generation sequencing (NGS) technology is now widely applied as a cost-effective approach to large-scale re-sequencing of many individuals from species where draft de novo genome assemblies are available. This advance offers unprecedented power to characterize genetic variation at the whole-genome level and provides important insights into the evolutionary forces that shape and maintain this variation. However, the utility of such NGS data rely crucially on the accuracy and quality of variant and genotype calling. Here, we provide a workflow for variant calling and genotype inference using a data set of whole-genome re-sequencing data of a number of European aspen (*Populus tremula*) individuals each sequenced to a depth of about 20 × coverage. We also estimate levels of nucleotide polymorphism and population differentiation of *P. tremula* using data from the first 500 scaffolds of twelve individuals sampled from four natural populations in Sweden. Compared to previous studies that were based on a small number of genes, we found higher genome-wide estimates of nucleotide diversity (π =0.00996) and lower genetic differentiation across populations (mean FST=0.00263).

INTER- AND INTRASPECIFIC GENETIC DIVERGENCE IN GENUS APODEMUS (RODENTIA, MURIDAE): A FRAMEWORK FOR NGS

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The genus *Apodemus* includes more than 20 species of field and forest mice, with some of them sharing similar morphologies and living in sympatry. Interspecific divergences were previously investigated by means of morphological, biochemical, karyological, and analyses of nuclear and mitochondrial gene sequences. The aim of this research was to describe the divergence among and within several *Apodemus* clades by comparing the structure of six microsatellite loci. Furthermore, the evolutionary distances among these clades were measured in the data sample of 1483 partial and complete cytochrome *b* sequences (GenBank accessions). Both microsatellite allelic diversity and polymorphism in presence and length data add to already established line of evidence on interspecific differences among *Apodemus* species. We strengthened the distinct position of subgenus *Sylvaemus*, and emphasized intraspecific diversity in *A. sylvaticus*. This intraspecific divergence in the presence of microsatellite loci was compared to divergences at the karyological level. Bearing in mind that a good phylogenetic framework contributes to the phylogenomic research, we pinpoint the optimal choice of sample size for next generation sequencing (NGS) and the populations of interest.

EVOLUTIONARY INFERENCES USING NUCLEOTIDE POLYMORPHISM AND DIVERGENCE DATA IN THE TOMATO CLADE

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The recent revolution in sequencing technology offers great prospects to answer some key questions in evolutionary biology, such as determining the main forces shaping patterns of nucleotide polymorphism within and between species. Due to their relatively recent divergence, differences in mating system, and the genomic resources available, wild tomatoes (Solanum section Lycopersicon) are ideally suited for the study of polymorphism and divergence at a genome-wide scale. Of particular interest is the relative importance of various evolutionary and ecological processes that have shaped these patterns, such as differences in mating system, demographic/speciation history, recombination, and natural selection. We have sequenced 104 transcriptomes of species-wide samples from seven green-fruited species. Based on ~1 million SNPs, we have characterized patterns of genome-wide nucleotide polymorphism and divergence between these species, which reflect varying proportions of ancestral polymorphism between them. Population-genetic analyses will assess the role of evolutionary forces and life history traits on these patterns. For example, levels of synonymous and nonsynonymous polymorphism and divergence will be used to disentangle the importance of purifying and positive selection in the context of varying effective population size, mating system, and recombinational landscape.

REPETITIVE DNA AND INSIGHTS INTO GENOME EVOLUTION IN INSECTS

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Simple sequence repeats are common features of insect genomes, considered to evolve mostly neutrally, with a high turnover due to high mutation rates. We studied the conservation of these repetitive DNA elements in evolutionary time scales in genomes of insect species and could show that large numbers of these microsatellites can be retained over many million years. We compared Dipteran species with Hymenopteran species in relation to divergence time, and found a loss of microsatellites twice as fast in the flies, indicating a slower genome evolution in Hymenoptera which is consistent with previous studies based on substitution rates in coding sequences. However, if the generation time is considered, the Diptera actually show an 8.5-fold slower rate than the Hymenoptera, suggesting a faster genome evolution. Such microsatellite loci conserved between genomes of related species furthermore allow for genome-wide high resolution analyses of synteny as well as the comparison, and improvement of genome assemblies. Interspersed repetitive elements correction Retrotransposons and DNA Transposons – are considered as mobile genomic "parasites" and can be found in most species. With a genome-wide scan in twelve bee species, we study the diversities and abundances of these elements. Whereas solitary bee species show high levels of both diversity and abundance, some lineages appear to have lost most of it. The eusocial bumblebee and stingless bee species lack many types of these elements, but the most extreme case are three analysed honeybee species with genomes that are almost devoid of any retrotransposons. Furthermore, we find evidence for potential horizontal gene transfer, indicated by high sequence homologies between transposon elements from very distant non-insect species. In general, our results show that repetitive elements in spite of enhanced mutation rates can be used as a powerful tool for analysing patterns of genome evolution.

GENOMIC BASIS AND POSSIBLE EVOLUTIONARY CONSEQUENCES OF ECTOSYMBIOTIC LIFESTYLE IN A SULFIDE-RICH ENVIRONMENT

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Ectosymbioses between invertebrates and chemosynthetic microbes are common and ecologically important in sulfidic marine environments such as hydrothermal vents, cold seeps, and organic-rich coastal sediments. In these habitats, invertebrates have developed stable ectosymbiotic associations with epsilon and gamma-proteobacteria. It has long been documented that Thiothrix, gammaproteobacteria belonging to the Thiotrichaceae family, are ectosymbionts of a marine amphipod. Nevertheless, the associations between gammaridean amphipods of the genus Nipharqus and sulfur oxidizing Thiothrix bacteria, discovered recently in the Frasassi caves of central Italy, were the first reported examples of chemosynthetic ectosymbioses from a freshwater habitat. The Frasassi caves are underground ecosystem solely sustained by chemoautotrophy. Mats of filamentous sulfur-oxidizing gamma- and epsilonproteobacteria cover the sulfidic cave water bodies. The Niphargus amphipods directly interact with these mats, but have established ectosymbioses with Thiothrix clades, which are furthermore distinct from Thiothrix of the free-living communities. With the advent of the nextgeneration sequencing technology, comparative genomics have been exhaustively used to study endosymbioses, but have so far never been applied to ectosymbioses. Our aim is to compare the genomes of Thiothrix ectosymbionts of Niphargus with those of free-living Thiothrix present in Frasassi microbial mats to uncover the genomic basis and possible evolutionary consequences of an Recent studies revealed that Niphargus-Thiothrix ectosymbioses are not ectosymbiotic lifestyle. restricted to Frasassi, but occur in subterranean environments across Europe, including Movile cave in Romania and Melissotrypa cave in Greece. These newly discovered associations, which involve different host species and *Thiothrix* ectosymbiont clades, may serve as natural biological replicates for our comparative genomic studies.

PHYLOGENOMICS UNRAVELS MAJOR EVOLUTIONARY TRENDS OF THE BASIDIOMYCOTA

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The Basidiomycota is the second largest group of fungi, encompassing a remarkably diverse and important array of life forms, from mushrooms to smuts and rusts. They play vital roles in recycling nutrients in the global environment and present serious threats to conservation and economic activities as pathogens and wood rotting species. The importance of these species allied with their relatively small and compact genomes have made them prime candidates for sequencing projects, generating a growing platform of genomic resources. However, due to the recent availability of these resources, there has still been no effort to use genome-scale data to study the evolutionary history of this group of organisms. Here we present the results of an ongoing phylogenomics project that includes not only a representative sampling of the Basidiomycota, but also of the sister phylum, Ascomycota, in order to produce a comprehensive data matrix amenable for testing several hypotheses about the evolution of the Basidiomycota. Using state-of-the-art orthology detection methods as well as established and custom made quality control pipelines, we were able to assemble several data sets that range from 614 putative orthologs across 47 species to 3093 putative orthologs across 114 species, including data from complete genome projects and EST databases. Regarding the species-gene ratio, this represents the most comprehensive data matrix assembled so far for the fungal kingdom, allowing us to produce resolved and highly supported phylogenetic trees even among the most deep relationships. Paradoxically, we have also obtained several incongruities between data sets, which may have a substantial impact on the current understanding of fungal evolution. Our current goals reside in achieving a better understanding of these incongruities and adding a geological time-awareness into the evolutionary history of the Basidiomycota. This work is being funded by project PTDC/AGR-GPL/119943/2010.

X AND Y CHROMOSOME DIVERGENCE IN THREESPINE STICKLEBACK: WHAT HAPPENS WHEN CHROMOSOMES CEASE TO RECOMBINE?

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In many species, selection pressures and fitness optima differ between the sexes. Heterogamy will facilitate this effect since sex chromosomes can harbor sexually antagonistic alleles. On evolving sex chromosomes, lack of recombination results in degeneration of the Y (or W) and may affect the speed at which beneficial alleles accumulate (faster X (or Z)). Recent evidence has demonstrated that chromosome XIX in threespine stickleback (*G. aculeatus*) is actually the nascent sex chromosome pair. The sex determining locus maps to this group and the Y contains various physical differences, including deletions, inversions and accumulation of repetitive DNA, indicating that the Y has started to degenerate. In order to determine the impact of degeneration on the function of genes residing on the Y, and hence a potential mechanism for the evolution of sex-specific traits, we examined genomic DNA and RNA transcriptomes from several individuals from two distinct lineages with independent Y chromosome populations since the last ice age. Since sex chromosomes and autosomes are presumed to evolve at different rates, we can examine the selective forces specific to the evolving Y chromosome. We used sequence capture and transcriptome sequencing to obtain sequence from the majority of the sex chromosomes as well as portions of several autosomes to predict the effects of Y degeneration on gene function in two geographically distinct populations: Alaska and Finland. When comparing the capture data with transcriptomes, over 68 000 variants were determined to be Y-specific. Many of the expressed genes from XIX in male are affected by nonsense and missense mutations or by mutations affecting splice sites. Despite the large amount of mutations, a large number of genes have a transcript from both X and Y, although incomplete splicing is also evident from the Y. If Y-mutations produce functional protein products, this could be a potential source of novel male-beneficial alleles.

INSIGHTS FROM SINGLE CELL GENOMICS AND METAGENOMICS INTO MICRODIVERSIFICATION PROCESSES IN FRESHWATER BACTERIA OF THE SAR11 CLADE

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The SAR11 clade include oceanic and freshwater alphaproteobacteria. SAR11 is one of the most abundant bacteria in the upper surface waters of the oceans. The freshwater group, referred to as LD12, is uncultivated. Comparison of closely related groups that inhabit very different environments give a unique opportunity to investigate mechanisms and forces that shape their evolution. Here, we analyzed single amplified genomes from ten LD12 cells. Phylogenetic analysis of concatenated pan-orthologous proteins confirmed presence of a monophyletic LD12 cluster within the broader SAR11 clade. Most of the single gene trees agreed in topology, with a few clear examples of incongruence due to recombination events. Oceanic SAR11 strains have among the highest estimated ratio of recombination to mutation in bacteria, but little is known about the LD12 clade. Our study indicated substantially lower recombination frequencies for LD12. Based on sequence shared between the nine most complete single cell genomes, calculated recombination to mutation ratio placed the LD12 at the opposite extreme of the scale compared to the marine sequences. Pairwise comparisons of substitution frequencies showed the presence of microclusters. The LD12 single cells also displayed conservation of gene order structure. The recruitments of metagenomic reads from six lakes in Sweden and the US were used to investigate phylogenetic representation of the single cells and estimate the population mutation and recombination rates. Clustering of metagenomic reads showed no relationship with the trophic status of these lakes, but indicated both geographic and seasonal variability. Some microclusters were present in several lakes and we hypothesize that periodic selection for adaptation to different ecological niches leads to variation in abundances depending on the lake and conditions.

CONSEQUENCE OF THE EVOLUTION OF SOCIAL BEHAVIOR IN SPIDERS ON THE INTENSITY OF SELECTION

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The evolution of social behavior in spiders has profound influence at most aspects the biology of the species. Especially, the selection intensity is predicted to be highly decreased due to low effective population sizes caused by common characteristics of severe inbreeding, female biased sex ratio, reproductive skew and strong meta-population dynamics. In the Stegodyphus genus three species have independently evolved social behavior. We compare historical selection intensities (dN/dS - interspecific) and current selection intensities ($\pi N/\pi S$ - intraspecific) among the three social species and their three solitary sister species to test the prediction of decreased selection intensities in social species. We have sampled 10 individuals from each of 5 populations in all 6 species, and sequenced ~1% of each genome (~30mb and ~300kb protein coding) using double digest RAD sequencing. We have recently sequenced the genome of one of the social species, which facilities the analyses. We find that effective population sizes are highly decreased in the three social species; however, current selection intensities are suggested to be highly reduced. These results suggest that social behavior evolved recently. The results are backed up by simulation studies of relation among $\pi N/\pi S$ and dN/dS.

EVOLUTIONARY DYNAMICS OF THE HUMAN NADPH OXIDASE GENES CYBB, CYBA, NCF2 AND NCF4: FUNCTIONAL IMPLICATIONS

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The phagocyte NADPH oxidase catalyzes the reduction of oxygen to reactive species with microbicidal activity. It is composed of two membrane-spanning subunits, gp91-phox and p22-phox (encoded by *CYBB* and *CYBA*, respectively), and three cytoplasmic subunits, p40-phox, p47-phox and p67-phox (encoded by *NCF4*, *NCF1* and *NCF2*, respectively). Mutations in any of these genes can result in chronic granulomatous disease, a primary immunodeficiency characterized by recurrent infections. By means of evolutionary mapping, we determined that episodes of adaptive natural selection have shaped the extracellular portion of gp91-phox during the evolution of mammals, which suggest that this region may have a function probaby in host-pathogen interaction. Based on a resequence analysis of ~35kb of *CYBB*, *CYBA*, *NCF2* and *NCF4* in 102 ethnically diverse individuals, we show that the pattern of diversity of *CYBA* is compatible with balancing natural selection, perhaps mediated by catalase-positive pathogens. *NCF2* in Asian populations also shows a suggestive pattern of diversity characterized by a differentiated haplotype structure. Our study provides insight into the role of a pathogen-driven natural selection in an innate immune pathway and sheds light on the role of *CYBA* in endothelial non-phagocyte NADPH oxidases, which are relevant in the pathogenesis of cardiovascular diseases and other complex diseases.

EVOLUTION OF HEME-COPPER OXIDASES SUPERFAMILY: RECONCILING GENE DUPLICATIONS, LOSSES AND TRANSFERS TO UNDERSTAND THE ORIGIN OF AEROBIC RESPIRATION

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To understand the origin and evolution of heme-copper oxydases, the terminal enzymes of aerobic respiratory chains, is fundamental to clarify the emergence of this important cellular process and how biological activity has shaped the history of our planet. Prior gene-phylogeny analyses were unable to clarify if they predate oxygenic photosynthesis already reducing oxygen, if they predate oxygenic photosynthesis but without reducing oxygen or if they postdate oxygenic photosynthesis. Using an enlarged sampling of 2723 genomes from 1485 prokaryotes (all available prokaryote genomes at November 2011) we characterized the distribution and number of the different types of HCO's across Archaean and Bacterial phylums. We further used a time-calibrated tree-of-life against which we reconciled the inferred HCO gene phylogeny, under duplication, loss and transfers models, to test hypotheses about possible evolutionary scenarios for this family. All different HCO clades seem to make equally parsimonious roots of the gene-tree and multiple hypotheses remain open as possible evolutionary scenarios, but overall, results point to an early origin and diversification of this genefamily, prior to the origin of photosynthetic algae, and also with high amounts of duplications and lateral gene transfers. Although difficult and computationally intensive, only the simultaneous consideration of different evolutionary phenomena acting at different evolutionary scales, such as duplications, losses, lateral gene transfers and incomplete lineage sorting may properly elucidate deep and shallow patterns of diversification and co-evolution.

SPECIES TREE AND POPULATION SIZE ESTIMATION OF IBERIAN AND NORTH AFRICAN PODARCIS

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Phylogenetic methods were initially developed having only a single genes and a single individual per species available. Recent technological advances provide us with data containing many loci and several individuals per species, which are essential to proper infer species diversification histories. We present a new maximum likelihood approach to estimate species tree and the gene trees based on the multi species coalescent (Rannala & Yang 2003). Our method accounts for incomplete lineage sorting and also enables us to infer the population sizes. We use simulations to compare our method with existing ones. And also analyse a dataset containing 20 species with about 200 individuals and around 16 genes of Podarcis wall lizards from the Iberian Peninsula and North Africa.

Symposium

18. Evolutionary Demography

23 and 24 August

Co-organised by:





Program

Friday 23 August

Session(s): 11, 12, 13

Saturday 24 August

Session(s): 14

Organisers: Ulrich K. Steiner and Barbara Pietrzak

Invited speakers: Rebecca Sear and Tim Coulson

Description:

This symposium aims to stimulate cross disciplinary exchange in the emerging field of evolutionary demography that explores evolutionary and ecological processes that shape birth and death of individuals and populations. Evolutionary theory is formulated in demographic characteristics including structure, fitness, selection, and change. Specific emphasis is given on aging and senescence with the individual serving as link between the lower mechanistic level and the higher functional level both from a biological and demographic perspective.

D23SY18IT14:00R5

HUMAN EVOLUTIONARY DEMOGRAPHY: ILLUSTRATED WITH REFERENCE TO THE IMPORTANCE OF KIN FOR HUMAN REPRODUCTIVE SUCCESS

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Evolutionary demographers working on our own species are fortunate: data on humans abounds, both from the real world (including large-scale national datasets collected by demographers and economists and data on small-scale traditional societies collected by demographers), and from the lab (psychological and medical). We also have access to the substantial amount of research done in the social and medical sciences on how to collect, analyse and think about such data. In this talk I will summarise the benefits of the cross-disciplinary approach of human evolutionary demography, which combines data, methods and insights from the social sciences with the theoretical framework of evolutionary biology. I will do this with particular reference to my research on kin influences on demographic outcomes, including child survival and fertility rates. As a social species, interactions with other individuals are important for human fitness. The 'cooperative breeding' and 'pooled energy budget' models of human social organisation suggest, in fact, that such interactions are essential for human reproductive success. Here I will present results from a comparative project which is investigating the empirical evidence that kin do matter for women's fitness across a wide range of human populations, including: the analysis of nationally representative datasets from both high and low income countries; a comparative analysis of datasets from traditional, subsistence societies contributed by anthropologists; and psychological experiments. This evidence demonstrates that the presence of kin is often correlated with higher reproductive success, but also that interactions between kin are not always necessarily cooperative. There is also some evidence for local resource competition between kin, and conflicts of interest between affinal kin (those related by marriage).

18. Evolutionary Demography

D24SY18IT10:30R5

USING EVOLUTIONARY DEMOGRAPHY TO LINK EVOLUTION AND ECOLOGY

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Tracking the dynamics of character distributions allows phenotypic evolution, life history evolution and the population ecology of a species to be studied simultaneously. In this presentation I will describe recent developments and applications of models that track such dynamics, and show how they have helped understand why rapid phenotypic and population change is so prevalent.

D23SY18RT14:48R5

EVOLUTION OF SENESCENCE IN HETEROGENEOUS LANDSCAPES

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The current theory of senescence is developed in a very simple ecological and demographic context, with a unique population at equilibrium in a homogeneous habitat. In the wild, species live in a variable environment in space and time, where the assumption of equilibrium is often transgressed. In this study, we use models of quantitative genetics in structured populations in order to investigate the evolution of senescence in a variable environment. Adaptation to local environment depends on phenotypic traits which expression varies with age. We study different scenarios where the environment changes abruptly, gradually or cyclically with time and where the environment is heterogeneous in space with different populations connected by migration. The strength of selection decreases with age, which predicts slower adaptation of traits expressed late in the life cycle, potentially generating stronger senescence in habitats where selection changes in space or in time. This prediction is however complicated by the fact that the genetic variance also increases with age. With numerical calculations, we found that in most cases the rate of senescence is enhanced when the environment varies. Especially, migration between different habitats is a durable source of senescence in heterogeneous landscapes. We also show that the rate of senescence can sometime decrease transiently, when the population is not at equilibrium, with possible implications in experimental evolution and in the study of invasive species. Our results highlight the need to study age-specific adaptation, as a changing environment can impact differently each age-class with different consequences on demography.

D23SY18RT15:12R5

MATERNAL-AGE EFFECTS ON RATES OF SENESCENCE DIFFER BETWEEN CLONES OF THE WATER FLEA, DAPHNIA PULEX: CAUSES AND CONSEQUENCES

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Senescence is widely believed to be the evolved consequence of life-history strategies that are themselves shaped by extrinsic mortality. Consequently, evolutionary explanations of senescence are static. But recent studies investigating lifespan extension, genetic intervention and dietary restriction suggest that rates of senescence are plastic and may be regulated by the same mechanisms that underpin life-history plasticity. The Lansing effect refers specifically to plasticity in lifespan that are derived from maternal age effects: offspring from older mothers often senesce at faster rates. Lansing demonstrated this effect in rotifers over 60 years ago, and Lansing effects have now been observed in a broad range of taxa. However, there is still no evolutionary framework for the Lansing effect, or a mechanism explaining how maternal age effects are transmitted and alter offspring life-histories. We repeated Lansing's experiments in D. pulex. Using offspring from the first clutch to set up young maternal lines, and offspring from the fifth clutch to set up old maternal lines, we reared three clones for three generations. Offspring development, life-history and rates of senescence were then compared in the fourth generation. Lansing Effects were observed irrespective of the measure of senescence used, and varied in strength between clones. Offspring from old maternal lines were born larger, grew more, initiated maturation at larger sizes, and had increased early lifetime reproductive effort. However, differences in the growth and maturation decisions of offspring from young and old maternal lines were size-independent, supporting Lansing's assertion that older mothers transmit a non-genetic "ageing factor" to their offspring that alters their life-histories. We propose a novel adaptive explanation for the Lansing effect, and discuss the implications that an interaction between genetic and non-genetic inheritance have life-history evolution and population demography.

D23SY18RT15:45R5

THE INFLUENCE OF NUTRIENT TYPE ON LONGEVITY AND REPRODUCTIVE SENESCENCE IN WILD AND LABORATORY POPULATIONS OF THE ANTLER FLY (*PROTOPIOPHILA LITIGATA*)

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Senescence has rarely been shown in systems with high rates of extrinsic (age-independent) mortality such as in wild insects. What is known about senescence is generally based on laboratory model organisms (i.e. exclude extrinsic mortality) or based on studies of wild populations that have not examined senescence experimentally. Thus, we have little information on the extent to which studies in the laboratory correspond to senescence in the wild. The observed patterns of ageing in these systems will be influenced by resource acquisition and allocation to various life-history traits. Theory predicts more rapid ageing in individuals with higher initial mating rates stemming from a trade-off in reproduction vs. somatic maintenance and repair. In turn, the proportion of various energy sources in food, or restriction thereof, may shift the balance in this life history trade-off through their influence on physiological state and condition. Moreover, high-profile effects of caloric restriction in increasing lifespan come from lab populations. Diet manipulations may have much more complex and sexdependent effects, especially in the wild. Here we take advantage of an extraordinary system - Antler Flies – where extreme site fidelity of males permits estimates of both lifespan and lifetime mating success. We extend previous work demonstrating aging in wild males to address the effects of a diet manipulation and to compare estimates conducted under lab vs. field conditions. Using a factorial design, we estimated senescence in mating rates and probability of survival for males treated with protein, carbohydrates, or water in both lab and field environments. We will discuss the extent to which these resources differentially influence life span, reproductive ageing, and the shape of survival curves. Our results have direct implications for the evolution of demographic patterns and senescence.

18. Evolutionary Demography

D23SY18RT16:09R5

GROWTH, MAINTENANCE AND REPRODUCTION IN ANT COLONIES

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Organisms have to allocate their available resources into growth, maintenance and reproduction throughout their life, and several life-history studies have documented a trade-off between growth and reproduction. However, the invested energy and costs for maintenance are usually difficult to evaluate in individual organisms. In this study, we monitor single-queen ant colonies of the species *Cardiocondyla obscurior* throughout their whole life. With this model organism, it is possible to not just measure the allocation of resources into colony growth/maintenance and into sexual reproduction but to gain insights in the time-dependent resolution of the allocation patterns throughout the complete life cycle of a large number of colonies and the consequences on colony fitness and queen lifespan. Specifically, we focus on investment into maintenance, which cannot be measured in whole organisms, and its consequences for subsequent colony survival.

D23SY18RT16:33R5

EXAMINING THE LINK BETWEEN GENOTYPE AND PHENTYPE: MITOCHONDRIAL GENE EXPRESSION LEVELS ACROSS DISTINCT HAPLOTYPES, SEXES, AND AGES

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Mitochondria are key components of cellular metabolic processing, providing most of the cellular energy required for survival. The small set of genes located within the mitochondria has recently been the subject of much attention by evolutionary biologists, as a groundswell of studies have documented that allelic variance within the mitochondrial DNA (mtDNA) often confers modifications to the phenotype. Mitochondria have been shown to play an active role in the process of ageing, as already proposed by Harman in 1972, and recent studies suggest that some of this allelic variance is even male-specific. Here we use the fruitfly Drosophila melanogaster as a model to advance our understanding of the link between the mitochondrial genotype and phenotype. We sequenced full mitochondrial genomes 13 lines and examine genotype-specific transcriptional profiles of 10 (out of 13) mitochondrial key genes on an isogenic nuclear background. Gene expression patterns are specifically examined for males and females and across age classes. Our goal is to understand the elusive mechanisms that mediate mitochondrially-encoded effects on the phenotype, and to home in on the candidate mutations that cause these effects.

D23SY18RT16:57R5

ONTOGENESCENCE AND THE BARNACLE: AN EXPERIMENTAL EXAMINATION OF EARLY LIFE MORTALITY IN THE ESTUARINE BARNACLE AMPHIBALANUS IMPROVISUS

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Ontogenescence, the decrease in mortality rate experienced during early life, is a nearly universal lifehistory trait. Among aquatic organisms this high and declining early mortality is generally attributed to extrinsic risks to which developing individuals gradually become more robust through increasing size and speed (Acquisition of Robustness). We examined the stage-specific mortality patterns of larval estuarine barnacle *Amphibalanus improvisus* in the laboratory, and found that in the absence of the usual environmental risks (predation, washing away, rapid environmental changes), early mortality was still high and generally declining, but was focused around developmental transitions. We further found that even though individuals of these transitional stages are more likely to encounter certain environmental stressors (temperature and salinity shocks), these stages exhibited the lowest tolerance to these stressors. Our results, while not refuting the Acquisition of Robustness hypothesis, lend support to the Transitional Timing Hypothesis, which states that ontogenescence arises because biological transitions are dangerous, and are concentrated early in life. Our data illustrate several ways in which mortality can be concentrated around early-life transitions.

D23SY18RT17:45R5

DEMOGRAPHIC HETEROGENEITY, SELECTION, AND POPULATION RESPONSE

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Demographic heterogeneity (unmodeled variation in traits underlying vital rates) has attracted much attention in recent years. Empirical studies show substantial heterogeneity in many populations. Theory shows that it can have strong impact on demographic variance (and by inference, extinction risk); some kinds of demographic heterogeneity can also have large effects on the mean population growth rate. Our studies of heterogeneity point to a strong connection with natural selection, and provide new insights on how changes in phenotype distributions can occur in structured populations. The key is an understanding of the within-population phenotypic correlation structure. Correlations can occur within individuals over time, between individuals at a given time, and between individuals at different times. In matrix models allowing persistent heterogeneity, survival heterogeneity increases the mean population growth rate, and the population becomes dominated by "good survivors" even if the parentoffspring correlation is zero or negative. By contrast, the effect of fertility heterogeneity depends on the sign of the parent-offspring correlation. This is because information transmission across time depends (for reproduction) entirely on the parent-offspring correlation, but for survival information is also transmitted by changes in the population's phenotype structure. With overlapping generations, then, selection on survival and on reproduction can cause different responses. The demographic response (change in stable phenotype distribution) occurs because of the non-genetic correlation structure of the population. For survival, this response occurs regardless of the parent-offspring correlation, but for reproduction, the response depends strongly on the parent-offspring correlation. This suggests that selection on survival may often lead to larger responses and faster changes than is the case for selection on reproduction.

D23SY18RT18:09R5

SHOULD I STAY OR SHOULD MY SIBLINGS GO? SIBSHIP EFFECTS ON DISPERSAL BEHAVIOUR

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Understanding dispersal behaviour and its determinants is critical for studies on life history maximising strategies. Though many previous studies have investigated the causes of dispersal, very few have focused on the importance of sibship, especially in humans. Using a large demographic historical dataset (n=4000), we investigate the influence of sibling competition and cooperation on dispersal behaviour. Specifically, we test whether the probability, the timing and the range of dispersal depend on the presence of same-sex or opposite-sex siblings and on their reproductive status. This study is the first to investigate in detail the importance of siblings' interactions for dispersal behaviour in humans and has important implications for the understanding of the evolution of family dynamics and fitness maximising strategies in humans and in other species.

D23SY18RT18:33R5

ESTIMATING FLUCTUATING SELECTION IN AGE-STRUCTURED POPULATIONS

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In age-structured populations, viability and fecundity selection of varying strength may occur in different age classes. On the basis of an original idea by Fisher of weighting individuals by their reproductive value, we show that the combined effect of selection on traits at different ages acts through the individual reproductive value defined as the stochastic contribution of an individual to the total reproductive value of the population the following year. The selection differential is a weighted sum of age-specific differentials that are the covariances between the phenotype and the age-specific relative fitness defined by the individual reproductive value. This enables estimation of weak selection on a multivariate quantitative character in populations with no density regulation by combinations of age-specific linear regressions of individual reproductive values on the traits. Demographic stochasticity produces random variation in fitness components in finite samples of individuals and affects the statistical inference of the temporal average directional selection as well as the magnitude of fluctuating selection. Uncertainties in parameter estimates and test power depend strongly on the demographic stochasticity. Large demographic variance results in large uncertainties in yearly estimates of selection that complicates detection of significant fluctuating selection. The method is illustrated by an analysis of age-specific selection in house sparrows on a fitness-related twodimensional morphological trait, tarsus length and body mass of fledglings.

D23SY18RT18:57R5

INDICATORS OF SELECTION PRESSURE, AND CHANGES IN VITAL RATES AT MULTIPLE AGES: A GENERAL METHOD

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Hamilton's (1966) indicators of selection pressure on age-specific additive changes in mortality and fecundity have provided evolutionary biology with the key to study evolutionary effects of age-specific changes in vital rates. However, other indicators of 'selection pressure', the sensitivity of fitness to some standardized perturbation of a vital rate, have been proposed and advocated, involving nonadditive changes in vital rates (Baudisch 2005), or changes that involve multiple ages (Hamilton 1966, Abrams 1991). In addition, it may be wearisome to obtain an indicator of selection pressure on more complex patterns of change across ages. Realizing that any eventual effect on fitness results not only from selection pressure, but also from the force of perturbation of the vital rates, we develop the calculus to evaluate the effect on fitness of the combination of selection pressure with a perturbation function. We apply this calculus to: 1) show that other indicators of selection pressure can be derived from Hamilton's 'elementary' indicators on additive perturbations; 2) show that choosing an indicator of selection pressure and the appropriate perturbation function is merely a matter of parameterization, rather than a true conceptual difference; 3) propose to use Hamilton's indicators of additive change for all analyses, since in this way all the biological variation is contained in the perturbation function; and 4) investigate in what direction a resident phenotype may evolve under some given life-history tradeoff that produces change in age-patterns of fecundity and/or mortality. Under 4), the central idea is that life-history trade-offs can be presented as perturbation functions that lead to a different perturbation (different magnitude and direction) at different ages. Implications for the evolutionary theory of aging are discussed.

18. Evolutionary Demography

D24SY18RT11:18R5

ACTUARIAL SENESCENCE INCREASES THE RISK OF EXTINCTION OF MAMMAL POPULATIONS

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Despite recent general acknowledgement that senescence has significant negative demographic manifestations in natural environments across a wide range of animal species, we still do not know if it can significantly impact the dynamics and viability of wild populations. Focusing on actuarial senescence (i.e., the decline of survival probabilities at old ages), we use specific demographic information to project the dynamics and extinction risk of wild populations of 58 species of mammals in the presence or absence of senescence. Our projections reveal major negative effects of ageing on population dynamics and viability, with a 27% decrease of the time to extinction on average and a potential deterioration of the species projected conservation status in 10% of the cases. The magnitude of the senescence cost is relatively homogenous among mammal orders at the exception of primates, which are disproportionally affected, due to their slow pace of life. Senescence is associated with particularly strong increases of the extinction risk in species with high annual survival probabilities of young adults and long intervals between litters, independently of their place in the phylogeny, indicating that the pace of life history can be used to forecast the detrimental effects of ageing on the viability of species.

D24SY18RT11:42R5

REDUCED LIFESPAN AND INCREASED AGEING DRIVEN BY GENETIC DRIFT IN SMALL POPULATIONS

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Despite recent progress in determining the genetic basis of ageing1-2, explaining the strong variation in lifespan among organisms remains one of the biggest challenges in evolutionary biology. Most previous work has concentrated on differences in selection regimes and selection pressures3-4, but evolutionary theories hold that ageing may evolve due to an accumulation of mutations with late-life deleterious effects via genetic drift5-6. As genetic drift also reduces genetic diversity within populations, a positive correlation between genetic diversity and lifespan is expected. Here we develop a model to formalize this idea. We then show that there indeed exists a strong positive relationship between lifespan and genetic diversity among populations of Daphnia magna and that ageing is accelerated in populations with relatively low genetic diversity (small populations). An experimental test provides compelling evidence that this correlation is due to increased drift in small populations rather than due to adaptation to environments that favour faster life histories: Inter-population crosses between small populations result in rescue phenotypes with lifespans as high as in large populations. Such rescue phenotypes are expected only if the short lifespans in small populations are maladaptive. Further confirmation comes from data on other life-history traits, which demonstrate a general increase in genetic load in small populations and clearly contradict the evolution of faster life histories. These results suggest that organismal lifespan and ageing can be strongly affected by genetic drift and can thus be maladaptive, especially in small populations where selection is less efficient.

POSTERS

D20SY18PS0509

AGE-SPECIFIC BREEDING SUCCESS IN A WILD MAMMALIAN POPULATION: SELECTION, CONSTRAINT, RESTRAINT AND SENESCENCE

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The Selection, Constraint, Restraint, and Senescence Hypotheses predict how breeding success should vary with age. The Selection Hypothesis predicts between-individual variation arising from quality differences; the other hypotheses predict within-individual variation due to differing skills or physiological condition (Constraint), residual reproductive lifespan (Restraint), or somatic and reproductive investment (Senescence). Studies tend to focus on either the initial increase in breeding success or later decrease; however, both require consideration when unravelling the underlying evolutionary processes. Additionally, few studies present genetic fitness measures, and rarely for both sexes. We therefore test these four hypotheses, which are not mutually exclusive, in a high-density population of European badgers Meles meles. Using an 18-year dataset (including 22 microsatellite loci) we show an initial improvement in breeding success with age, followed by a later and steeper rate of reproductive senescence in males than in females. Breeding success was skewed within age-classes indicating the influence of factors other than age-class. This was partly attributable to selective appearance and disappearance of badgers (Selection Hypothesis). Individuals with a late age of last breeding showed a concave down relationship between breeding success and experience (Constraint Hypothesis). There was no evidence of abrupt terminal effects; rather, individuals showed a concave down relationship between breeding success and residual reproductive lifespan (Restraint Hypothesis), with an interaction with age of first breeding only in females. Our results demonstrate the importance of investigating a comprehensive suite of factors in age-specific breeding success analyses, in both sexes, in order to fully understand evolutionary and population dynamics.

D23SY18PS0046

AGEGUESS CROWDSOURCING HUMAN AGING RESEARCH

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Human lifespan increases by 2.5 years every decade. This unprecedented change in life histories poses fundamental challenges for evolutionary theories. Some evidence indicates that this change is due to a delay in aging rather than a change in the rate of aging. AgeGuess is a citizen science project and online game that investigates the differences between perceived age (how old you look to other people) and chronological age (how old you actually are) and their potential power as an aging biomarker. Is the increased life expectancy reflected in how old one looks, i.e. are the new 60's the old 50's? Are people who look older than they are more likely to die early? Does the rate of looking older differ among individuals or some individuals just looking older all their lives, i.e. does the difference between estimated and real age change over time? Is the difference between perceived and real age heritable? Are there periods in life when one ages faster? The project aims at such questions by a simple on-line game in which you can post your photos, have other people guess your age, as well as guess the age of other users. Curious? Please visit AgeGuess.org

18. Evolutionary Demography

D23SY18PS0089

IS THE X-CHROMOSOME A HOTSPOT FOR GENES AFFECTING SEX DIFFERENCES IN LIFESPAN AND AGEING?

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This project (currently ongoing) aims to estimate the individual contributions of the X-chromosome and major autosomes to variance and sex differences in lifespan of *Drosophila melanogaster*. We test the hypothesis that the X-chromosome should be a hotspot for genetic variation causing sex differences, in particular we test the contribution of the X and autosomes to sexual dimorphism and sex specific variance in lifespan and ageing. Further to this we are also testing contributions of the Y chromosome to variance in male lifespan and ageing.

WHY DO THE WELL-FED DIE YOUNG? AN ECO-EVOLUTIONARY PERSPECTIVE ON THE EFFECTS OF DIETARY RESTRICTION

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The phylogenetically conserved life-history responses to dietary restriction (DR) – lifespan extension and reduced fecundity – have long been thought to reflect an adaptive survival mode whereby resources are redirected from reproduction to somatic maintenance and repair, enabling organisms to weather periods of resource scarcity. We argue that this interpretation of DR's effects is both inconsistent with recent experimental evidence and at odds with the ecology of natural populations. Laboratory studies have shown that high nutrient intake stimulates nutrient-sensing pathways that enhance reproduction as well as the ability to respond to environmental challenges, but impose latent costs -- increased rates of cellular damage and aging – that reduce survival in the lab. However, relative to laboratory organisms, natural populations subject to high extrinsic mortality rates are far less likely to reap the survival benefits associated with DR, or to pay the fitness costs associated with full-feeding. We suggest that DR's effects are best understood as the corollary of an ancient and highly-conserved strategy that allows organisms to take advantage of periods of resource abundance.

THE ECOLOGY OF SEXUAL CONFLICT: PARALLEL EVOLUTION OF MALE HARM AND FEMALE RESISTANCE IN EXPERIMENTAL POPULATIONS OF DROSOPHILA MELANOGASTER

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Sexual conflict arises when divergent reproductive strategies of males and females generate sexually antagonistic selection that can have large impacts on mortality and reproductive output of both sexes. Sexually antagonistic selection is also recognized as an important driver of divergence and speciation, as male-female arms races may escalate in different phenotypic dimensions in separate populations, even in the absence of environmental differences. However, ecology can have large impacts on the evolution of sexual traits, and can even promote their parallel evolution. Environmental selection may therefore constrain the pathways of sexually antagonistic coevolution, although ecology has not previously been considered in investigations of sexual conflict. We provide some of the first evidence illustrating the importance of environmental selection on sexual conflict, using experimental populations of Drosophila melanogaster adapted to two different environments. Contrary to previous theory, we find no evidence that females have highest resistance to the harm of coevolved males. However, we do find evidence for parallel evolution of male harm and female resistance to male harm, as measured through female lifespan and senescence rates, associated with adaptation to these alternative environments, suggesting that sexual conflict may be predictable based on ecology. Ecologically-induced parallel evolution of traits under sexual conflict may have large impacts on how populations adapt to new environments, and how new species are formed, and ecology must therefore be integrated into considerations of sexually antagonistic coevolution.

GROUP FORMATION AND INDIVIDUAL MORTALITY

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In some species, organisms perform key activities in groups. This is the case, for example, when nesting or foraging is communal. The evolutionary advantage of this sort of social behaviour is that group members gain access to vital resources in an amount greater than that accessible through individual effort. In this paper, we explore the relationship between group formation and individual mortality in social species. We make two realistic assumptions: (i) there is a time lag between group formation and the moment in which group members enjoy the result of group activities (i.e. laborious construction of the communal nest, human agriculture); (ii) one or more group members may die during this time interval, and remaining members benefit from this event (i.e. the result of group activities is divided among fewer individuals). Two different situations are considered. First, mortality is independent of age, initially. We examine whether changes in individual mortality may have an impact on fitness when grouping is random. Second, mortality increases with age; i.e. ageing. In this situation, the strategy of choosing older individuals as group mates is selectively superior to random grouping. However, selection on "go-for-the-old" appears to be frequency dependent. Finally, we discuss the potential advantages of "segregate-by-age", a behaviour that has been observed in some species.

THE INFLUENCE OF POPULATION SIZE ON HOST-PARASITE COEVOLUTION

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Host-parasite interactions are ubiquitous in nature and have a strong impact on species evolution. The signatures of this impact have been identified in genomes, natural communities and on a phylogenetic level. It is not surprising that many aspects of host-parasite relationships have received particular attention from evolutionary biologists. Paradoxically, one indispensible and basic property of hostparasite interaction, population size oscillations, has been overlooked as a factor in host-parasite coevolution. Parasites, by reducing host fecundity and survival, strongly affect population size of the host, which very often is their only ecological niche. Already in the 1920s Lotka and Volterra showed that antagonistic interactions between species would lead to interdependent oscillations in their population size. However, most of the current models of host-parasite coevolution ignore population size changes or use a deterministic approach which cannot realistically imitate the finite nature of real populations. Similarly, in most experimental studies on host-parasite coevolution the population size is kept constant as a matter of good practice. To enhance a more realistic understanding of the coevolutionary dynamics, we performed laboratory-controlled evolution experiments with the model nematode host Caenorhabditis elegans and its microparasite Bacillus thuringiensis and specifically varied the factor population size. Here, we will show our results on temporal changes in host fitness and parasite virulence under different population size regimes.

REPRODUCTION AND LIFESPAN: ONLY AGE AT LAST REPRODUCTION MATTERS

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Reproduction is costly for mothers due to energetic and physiological requirements during pregnancy, lactation and childcare. Fathers are usually neglected by the studies since they do not pay direct costs of reproduction. However, including fathers in analysis is crucial, since it could suggests explanations about the mechanisms responsible for the relationships observed for mothers. Our study investigated how reproductive history is related to parental lifespan. We present results from historical Polish population belonging to the area of Mogielica Human Ecology Study Site. Demographic data for marriages that occurred from the year 1782 to 1882, collected from parish records were used. Relationship between number of children born, age at first reproduction and last reproduction, mean interbirth interval and parental longevity was analyzed controlling for year of birth, spouse's longevity and first birth interval (as an indicator of energetic status at the beginning of reproduction for women). Moreover, sex of the last child was included in the model to control for maternal condition at the end of reproduction (since sons are more costly than daughters). In separate models, number of children that survived to adulthood was analyzed. Both maternal and paternal lifespans were affected only by the age at last reproduction. The later the last child was born the longer lives parents had (mothers: HR=0.96; CI=0.93-0.99 and fathers: HR=0.95; CI=0.92-0.98). Similar relationships were found when number of children who survived to adulthood was analyzed (mothers: HR=0.96; CI=0.94-0.98 and fathers: HR=0.95; CI=0.93-0.97). The results suggest that in studied population there are no trade-offs between costs of reproduction and lifespan. Only the age at last reproduction is significant predictor of length of life. However, the fact that the same relationship is observed for fathers suggests importance of social rather than biological factors.

EXPLORING VARIATION IN LONGEVITY: TRANSCRIPTOMIC STUDIES IN LONG-LIVED ANT QUEENS

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Senescence occurs in all multicellular and unicellular organisms. It is generally assumed that lifespan is genetically determined. Several candidate genes are suspected to be responsible for a gradual deterioration of physical function. These findings mainly arise from experiments with rather short-lived organisms (flies, worms, mice). However, the enormous intraspecific variation in longevity is often disregarded. Social insects with their different castes (queens, workers, males) have plastic life history traits which also include different aging rates. Ant queens are famous due to their exceptional long life spans compared to workers and males, making them a good model for the study of aging. The myrmecine tramp ant *Cardiocondyla obscurior* lacks the "reproductive senescence" which is normally present in other organisms (Heinze & Schrempf 2012). In addition, female-male co-evolution determines the lifespan of the queen (Schrempf et al. 2005, Schrempf & Heinze 2008, Schrempf et al. 2011). However, even queens mated with a sterilized male live considerably longer and start to lay eggs earlier than virgin females. In order to gain insights into lifespan-mediating mechanisms of mating, we conducted RNAseq of gene expression of old queens that were subjected to different mating. By comparing old queens mated to a fertile or a sterile male with virgin queens, we aim to find genes involved in the regulation of queen longevity.

MODELING STEM CELL DYNAMICS TO UNDERSTAND NEGLIGIBLE SENESCENCE IN HYDRA

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Unlike other metazoans, *Hydra* do not undergo the distinctive rise in mortality with age known as senescence, resulting from an increasing imbalance between damage and repair. We propose that *Hydra* control their damage accumulation mainly by damage-dependent cell death and cell sloughing. We examine our hypothesis with a model that combines cellular damage with stem cell renewal, differentiation and elimination. *Hydra* individual can be considered as a large single pool of three types of stem cells with some features of differentiated cells. This large stem cells community prevents "cellular damage drift", which is inevitable in complex conglomerate (differentiated) metazoans with numerous and generally isolated pools of stem cells. The process of cellular damage drift, similarly to the process of genetic drift, is based on changes in distribution of damage among cells due to random events. In the model events that are sources of randomness include: budding, damage caused by cellular death, and undergoing and repairing cellular damage. Results favor the view that non-senescence is possible only in simple *Hydra*-like organisms characterized by high proportion and number of stem cells, high cell division rate, an effective cell selection mechanism, and having stem cells with the ability to take some roles of differentiated cells.

MODELING DEMOGRAPHIC RISK DUE TO DISTANT INTERSPECIFIC HYBRIDIZATION

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Interspecific hybridization is frequent in nature and may deeply impact species evolution. This process may be enhanced by human activities, such as translocation of invasive species or habitat modification, and may consequently threaten species persistence. Hybridization between distantly related species often leads to infertile hybrids or fertile hybrids without chromosomal recombination during gametogenesis. Here, we present an original model describing those types of hybridization, which also considers density-dependent competition, assortative mating and dominance/recessive inheritance. Using our model, we assessed the demographic risk due to interspecific hybridization in two case studies. We first model the asymmetrical reproductive success between Atlantic salmon (Salmo salar) and brown trout (Salmo trutta), whose hybridization has been increased recently due to releases of domestic fishes in the wild. Second, we study the impact of the invasive frog Pelophylax ridibundus on two western European endemic waterfrogs, Pelophylax lessonae and Pelophylax esculentus, when introduced from different geographical origins having singular hybridogenetic properties. The results of our studies give the conditions under which interspecific hybridization can lead to population extinction. In both examples, the threat is demographically mediated, either through wasted mating potential or through demographic displacement of species. Our results highlight why distant hybridization should be considered in conservation assessments, and provide clues for conservation actions

UNCOUPLING INDIVIDUAL INTRINSIC QUALITY FROM HABITAT PREFERENCE AND QUALITY IN A TERRITORIAL SONGBIRD

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Theory predicts that animals breeding in heterogeneous landscapes will preferentially occupy habitats that maximize individual fitness, but identifying those habitats is problematic. Many studies develop metrics of habitat quality, but few identify the independent effects of individual animal 'intrinsic quality' on fitness from the effects due to the quality of the habitats they occupy. Several population processes also exist which may limit the degree to which individual and habitat quality are positively related in nature. We show that in an insular population of song sparrows (*Melospiza melodia*), females nested preferentially in breeding sites with high mean reproductive output over 35 years of study. Variation in site-specific reproductive output was positively related to female intrinsic quality, measured here as relative lifetime reproductive success (LRS). Vegetation characteristics predicted breeding site preference but were not closely linked to site-specific variation in annual reproductive output. Our results suggests that although females with high relative LRS exhibited no pattern in their occupation of more- and less-preferred habitats, the quality of individual females accounted for a large fraction of observed variation in the reproductive output of individual breeding sites. Thus, the intrinsic attributes of individual females influenced our estimates of what constituted 'higher-quality habitat'. Our results imply that estimates of the genetic and permanent environmental effects on individuals may be necessary to reliably assess habitat quality independent of its occupants, and avoid misrepresenting the potential effects of different habitats on animal fitness.

MOTHER-DAUGHTER CONTRACT AND AGE AT MENOPAUSE

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Age at natural menopause is characterised by significant variability, but the factors responsible for this variation are still not well recognised. Humans are cooperative breeders and non-reproducing females play important roles in raising children. Human females exhibit an extraordinarily low level of generational reproductive overlap, much lower than is the case for females of other primates, to minimize reproductive competition between generations over resources. We propose an evolutionary "mother-daughter contract" hypothesis that suggests that the oldest daughter helps her mother to raise vounger siblings but, in return, expects her mother to cease reproduction, shifting energy and time once her daughter's children are born. Data were collected by a questionnaire from 914 Polish postmenopausal women. 506 women, 44 to 98 years old, who had at least one child and who went through a natural menopause were included in the analysis. A woman's age at menopause was sensitive to the age at which she had her first daughter. The age of giving birth to the first daughter, even when she was not her first child, positively correlated with the age of the mother's menopause (N=332, P<0.02), while the age of giving birth to a first son did not have a statistically significant effect (N=332, p=0.36). Results of our study suggest that research on the menopausal transition should take into account mother-daughter relationships as potentially important determinants of the timing of menopause. The existence of the proposed mother-daughter contract is supported by research indicating an important role of older daughters and non-reproducing maternal grandmothers in raising children. We predict that even stronger correlation would be present between a woman's age at menopause and the age when her oldest daughter starts her own reproduction.

A COMPARISON BETWEEN HERITABILITIES OF LIFE HISTORY AND MORPHOLOGICAL TRAITS IN HUMAN POPULATIONS

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According to Fisher's fundamental theorem of natural selection, the amount of additive genetic variance in a trait, and hence its heritability, should decrease as the strength of selection on this trait increases. Supporting this idea, many authors have reported lower heritability values in life history traits than in morphological and physiological traits for different species. But there are no studies comparing the evolvability of different types of traits in human populations. The pedigree collection of decorated skulls from the historical population of Hallstatt (Austria) offers an exceptional opportunity to compare the heritabilities of life history and morphological traits in humans. In this study we first used church records to reconstruct the pedigrees and to obtain the values of individual life history traits, such as fertility, age at first and at last child, mean interbirth interval, adult lifespan and lifetime reproductive success). Second, we measured a sample of 353 complete adult skulls falling into the pedigreesusing a 3D Microscribe digitizer. A set of 50 landmarks were measured on each skull and from the 3D landmarks coordinates we estimated several size and shape variables reflecting the complex and modular structure of the human skull. We estimated the heritabilities of the life-history and morphological traits using a Restricted Maximum Likelihood method and statistically compared the resulting heritability values. On average, our results show lower heritabilities for life history traits than for morphological ones, confirming the initial hypothesis.

INFERENCE OF THE DEMOGRAPHIC HISTORY OF DROSOPHILA SUBOBSCURA USING APPROXIMATE BAYESIAN COMPUTATION: A MULTILOCUS ANALYSIS ALONG CHROMOSOME J

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Understanding the forces that control genetic variation in natural populations has been a major challenge for evolutionary biologists. *Drosophila subobscura* is a member of the obscura group that is widely distributed in the Paleartic region and presents a rich chromosomal polymorphism. Although this species has been extensively studied in relation to chromosomal inversion polymorphism, little is known about its demographic history. Multilocus studies provide an excellent opportunity to determine whether demographic factors have shaped the genetic variation observable in natural populations. In order to infer the demographic history of *D. subobscura* from the Paleartic area through Approximate Bayesian Computation (ABC), we sequenced and analyzed 16 non-coding regions distributed along the J chromosome in regions not affected by chromosomal inversions. In the 16 regions, the frequency spectrum was shifted towards an excess of low frequency variants, which led us to explore different simple scenarios including a final population expansion. Even if scarce, existing information on climatic changes as well as on recombination rates in the species under study was used to establish prior distributions for those parameters defining each model. Contrasting the different models through ABC analysis will provide new insides into the evolutionary history of *D. subobscura* based on an ample dataset of independently evolving loci not affected by chromosomal polymorphism.

COOPERATION AND CONFLICT IN HUMANS IN TRADITIONAL LARGE JOINT FAMILIES

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Group living can be associated with cooperation and even cooperative breeding whereby nonreproductive individuals help to raise offspring that are not their own. However, it can also lead to evolutionary conflict, which is a less studied phenomenon. Humans are considered to be cooperative breeders, since mothers commonly gain help in raising offspring from other (usually related) group members, such as grandmothers and siblings. Nevertheless, simultaneous breeding in the same household among reproductive-aged females, such as mothers-in-law and daughters-in-law, has also been linked with reduced success. The importance of cooperation and conflict is likely to vary according to ecology and social structure of populations, leading to differential selection pressures on dispersal patterns. We used life-history data on humans collected from church book records from 19th century Eastern Finland where joint-families were traditionally common. In joint families several adult offspring, usually sons, stayed in their natal farm with their families. This creates a situation where reproductive-aged women are not related to other women in the family, leading to possible conflict over resources and lowered fitness, but cooperation between women is also possible. We analyse mother's fecundity and survival of her offspring in relation to the presence and reproductive history of other reproductive-aged women in the family by event history analysis, whist controlling for potential confounders such as presence of other family members and temporal variation in mortality and fertility rates. Preliminary analysis suggest that living in larger joint families was beneficial for women's fitness, both in terms of fecundity and offspring survival, compared to smaller nuclear families. These results suggest that in this population cooperation between family members was more important than conflict, potentially favouring reduced dispersal among adult siblings

VARIATION IN COLONY TYPE DRIVES DEMOGRAPHY IN A RED ANT

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The demography of a population depends on the life history of individuals: when they reproduce, how many offspring they have, and how the offspring disperse. However, ants live in colonies and important life-history decisions are made at the colony level. It is known that the social organization of colonies influences the demography of ant colonies, e.g. the life of colonies headed by a single queen follows the lifespan of the queen while supercolonies (headed by an extremely high number of queens) can in principle live forever due to queen replacement. Here we studied the effect of different social environments on life-history traits at the individual, colony and population level in a single population of the common red ant (Myrmica rubra). We sampled ant workers and reproductives from 42 nests inside and outside a supercolony, respectively, for morphological and population genetic studies to compare important life-history characteristics such as body size, dispersal ability, resource allocation and developmental stability. Local density of ants around the nests was also measured. Our results show that very high density, extremely high effective queen number (>100) and moderately high inbreeding (F = 0.166) characterize nests in the supercolony. Surprisingly, virgin queens were produced exclusively in nests outside the supercolony having few queens (1.22), low inbreeding (F = 0.082) and low local density. In contrast, nests outside the supercolony that did not produce virgin queens were highly inbred (F = 0.39) and had high, but not extreme queen numbers (14.9). We hypothesize that inbreeding affects the future of diploid virgin queens but not haploid males, so that colony sex ratio may correlate with inbreeding. These findings together with our further morphological and molecular studies shed light on how large and long-lived ant supercolonies might evolve and are maintained in interaction with small but queen-producing colonies of the same population.

THE IMPACT OF AGE-RELATED CHANGES IN FERTILITY: IMPLICATIONS FOR SEXUAL CONFLICT

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The impact of age on evolutionary processes is a relatively under-appreciated consideration in biology, despite differential effects on reproductive physiology between females and males. It is not expected that fertility will decline evenly between the sexes, since different traits determine fertility in each sex. Using a large, pedigreed laboratory population, we conducted a quantitative genetic study of fertility in a common European rodent, the bank vole (*Myodes glareolus*), to explore how senescent changes affect coevolution between the sexes. Our results indicate that reproductive success, as measured by litter size in females and offspring sired in males, differentially declined according to sex, with females experiencing a sharper decline in their fertility than males. The genetic correlation between the sexes for fertility was not significant for younger individuals, but became significantly negative for older individuals that reproduced. The age-dependent expression of fitness-related genes results in a divergence of reproductive interests between the sexes. Furthermore, the additive genetic variance for female fertility increased with age, consistent with the process of relaxed selection in older age. These results will be discussed in the broader context of sexual conflict theory.

EFFECTS OF CLIMATE ON SURVIVAL OF ASIAN ELEPHANTS

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Climate change has intensified interest in understanding how climatic variability affects animal life histories. Despite this, little is known of their effect on survival in those species. Asian elephants (Elephas maximus) are endangered across their natural distribution, and inhabit regions often characterised by high seasonality of both temperature and rainfall. We investigated the effects of monthly climatic variation on survival and causes of death in Asian elephants of all ages and both sexes, using a unique demographic dataset of 839 semi-captive longitudinally monitored elephants from four sites in Myanmar between 1965 and 2000. Temperature had a pronounced effect on survival, with the lowest predicted survival during the hottest and coldest months in both sexes across all ages. Because during a year the elephants spent twice as long in temperatures higher than their optimum (240C) rather than temperatures below it, most deaths occurred during the "too hot" rather than the "too cold" period. Decreased survival at higher temperatures resulted partially from increased deaths from heat stroke and infectious disease, whilst the lower survival in the coldest months is associated with an increase in non-infectious diseases or poor health in general. Variation in survival was also related to rainfall with the highest survival rates during the wettest months. Our results show that even the normal-range monsoon variation in climate can exert large impact on elephant survival in Myanmar leading to large absolute differences in mortality, particularly among the youngest age classes. The persistence of a long-term trend towards higher global temperatures combined with the possibility of higher variation in temperature between seasons may pose a growing challenge to the survival of species such as the endangered Asian elephants.

EVOLUTIONARY DEMOGRAPHY OF MICROBES: GROWTH, SEX AND DEATH IN A MARINE PLANKTONIC DIATOM

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Populations of unicellular microalgae are orders of magnitude larger than those of multicellular organisms. They reproduce clonally by binary fission producing massive and, at times, periodical blooms. They cope with dramatic loss factors, such as primary consumers, parasite or viral infections. A sexual phase has been also reported for many species. Sex has a high cost in short-term, since cells have to stop division and invest energy in meiosis. Nonetheless, sex has been conserved in many microalgae, since it allows clonal populations to escape extinction, thus reducing its evolutionary cost in the long-term. Among unicellular microalgae, diatoms are key-players in marine ecosystems. Their landmark is represented by the rigid box-shaped siliceous cell-wall that imposes a progressive decrease in the average cell size of the cell population undergoing mitotic divisions. Larger cell size is generally restituted by sexual reproduction that, in diatoms, combines genetic recombination with the production of 'young' large cells. We have chosen the diatom *Pseudo-nitzschia multistriata* as a model to explore the application of a demographic approach to a planktonic microalga at the LTER station in the Gulf of Naples. The approach included in-situ monitoring of cell abundance and cell size, in-vitro experimentation on the factors that induce the transition between clonal reproduction and the onset of the sexual phase, and modeling simulations. We could follow the birth, maturation, and disappearance of various 'age classes' in the natural environment where large-sized cells - indicative of sexual reproduction events - were detected every two years. Our data suggest a collective behavior, apparently fostered by quorum sensing mechanisms, that shapes the periodic alternation between asexual and short-term sexual phases. Finally, our study corroborates the possibility to apply an evolutionary demographic approach to the study of unicellular organisms.

DISPOSABLE SPRINGTAILS: HIGHLY PLASTIC AGEING PATTERNS ARE EXPLAINED BY RESOURCE ALLOCATION TRADE-OFFS IN FOLSOMIA CANDIDA

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Although often neglected, the evidences of senescence occurring in the wild are accumulating (1) and there is a growing interest towards a clarification of how the mortality trajectories have been shaped by the ecological conditions (2). The challenge now lies in understanding the diversity of ageing patterns in the light of evolutionary theory (3). We developed an experimental system in the laboratory to assess these questions on the small and long-living Collembola Folsomia candida (4,5). We performed long term microcosms experiments to question how mortality trajectories have been shaped on the shortand long-term by trade-offs between traits. We found that within species genetic differences in ageing patterns can be explained with differences in growth and reproductive strategies: comparison of different lineages showed that initial mortality rate and age at onset of senescence are negatively correlated - a result coherent with recent predictions based on the 'disposable soma' theory of ageing (6). We also demonstrate that plastic adjustments of major life history traits triggered by changes in resource availability even late in life lead cohorts to shift from constant mortality trajectories (negligible senescence) to accelerated senescence. Our results emphasises the need for a more integrated ecological comprehension of the effects of environment and its fluctuations to understand how natural selection shapes ageing patterns. 1. D. H. Nussey et al., Ageing Research Reviews 12, 214-25 (2013). 2. A. Baudisch, J. W. Vaupel, Science 338, 618-9 (2012). 3. M. Bronikowski et al., Science 331, 1325-8 (2011). 4. T. Tully, R. Ferrière, PLoS One 3, e3207 (2008). 5. T. Tully, A. Lambert, Evolution 65, 3013-20 (2011). 6. M. J. Wensink et al. Biogerontology 13, 197-201 (2012).

STUDYING THE EMERGENCE AND EVOLUTION OF APOPTOSIS USING EVOLUTIONARY GAME THEORY AND AGENT BASED MODELLING

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Both antagonistic pleiotropy and disposable soma theory have predicted senescence (and in turn apoptosis) to be a `side effect'. Although the altruistic benefit of apoptosis has been suggested before, we are attempting to show that in a resource restricted environment, apoptosis can be a strategic choice. To demonstrate this, we employ a theoretical model based on Game Theory (GT) and computer simulations using Agent Based Modelling (ABM). It has been found that genetic mutations can transform the cells in a healthy tissue into a population of individualistic tumour cells. From a theoretical point of view, non-apoptotic cells can also become apoptotic. This allows us to start with a population of non-apoptotic cells and allow some of the cells to randomly mutate and evolve an apoptosis machinery. All these cells age, which will affect the cell's fitness, and may die due to environmental causes. We use a GT model to govern the strategies and payoffs. Initially the cells and the nutrients are randomly placed in the lattice and the cells are allowed to migrate to nearby lattice spaces by paying the cost of migration. The cells compete for food, which is then allocated according to the GT model. Once a cell secures enough nutrients, it replicates and the newborn is added to the population. After each time-step the nutrients are replenished. The non-apoptotic cells have a choice to become apoptotic and vice versa. The simulation is repeated over many time-steps and by varying parameter values. We find that switching to apoptotic mode gives the cell an advantage over nonapoptotic cells in a resource restricted environment. Analysis using Evolutionary GT indicates that apoptosis is an Evolutionary Stable Strategy (ESS) as long as the conditions remain the same. We also demonstrate how the model could potentially improve the disposable soma theory in relation to chronological lifespan. The proposed model has some features that may be of interest to mathematical oncology.

PERCEIVED PREDATION RISK ACCELERATES AGING IN WATER FLEAS

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Exposure to chemical cues of fish triggers adaptive response in water flea life history, leading to increased early reproduction. This has a cost, as lifespan in the environment free of such cues of predation is 20% longer. The aim of this study was to establish the biodemographic basis of phenotypic differences in water flea lifespan that are induced by the cues of fish predation, and to test for the change in the rate of aging, in particular. We examined mortality using large heterogeneous cohorts of two cladoceran species, *Daphnia longispina* and *Diaphanosoma brachyurum*, and found that the exposure to these chemical cues increase the Gompertzian rate of mortality acceleration. The phenotypic response is comparable to previously shown genetically based differences between *Daphnia* from habitats of different predation risk. The cue of extrinsic mortality risk from fish predation – a key factor shaping cladoceran life histories in the wild – is one of the few interventions shown so far to induce a plastic change in the rate of aging.

MISMATCHING THE MEDFLY: DEMOGRAPHIC EFFECTS OF DIETARY CARBOHYDRATES IN CERATITIS CAPITATA

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When environments vary over time and space individuals can be 'mismatched' - in an environment to which they are not adapted. In the short term, individuals may show plastic responses, which can buffer deleterious effects. Given sufficient evolutionary time and selection, organisms will also adapt genetically. The thrifty phenotype hypothesis considers plasticity to the prevailing environment. During development, the likely quality of the environment into which the individual will emerge is 'assessed'. Thus metabolic traits such as insulin sensitivity are plastic and can be 'set' during development. However, if the environment changes rapidly, the organism is set for the wrong environment, with deleterious consequences. This study considers the response to rapid changes in environmental quality using the widely successful polyphage Ceratitis capitata, the Mediterranean fruit fly. By altering the content of carbohydrate in the larval and adult diet, we created a mismatch scenario in order to test the impact of this rapid change on longevity as well as overall male and female fitness. The results of this study are discussed in terms of their relevance to the generalist nature of this species, as well as their consequences in terms of adaptation and sexual selection.

Symposium

19. Evolutionary Consequences of Deception

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3

Organisers: Carita Lindstedt-Kareksela and Mikael Mökkönen

Invited speakers: Martin Stevens, Tom Sherratt and Johanna

Mappes

Description:

The ability to deceive oneself, conspecifics or individuals of other species is a fundamental aspect to many coevolutionary struggles. Brood parasites have the ability to produce eggs that exactly resemble the egg coloration of their host species, while alternative mating strategies employed by 'sneaker' individuals circumvent confrontation in intrasexual competition - these are just a few examples of widely known deceptive strategies in the animal kingdom. Deception allows individuals to gain an evolutionary advantage in reproduction and/or survival, such as through deceptive colour mimicry, predator-prey systems, sexually antagonistic encounters with a mate, parent-offspring interactions or competition over resources. Much of the existing research on deception has been fragmented into various topics with limited interaction, even though there are some common themes such as frequency dependent selection, manipulation of the receiver's sensory system, and the antagonistic coupling of the actor's benefits to costs of the recipient(s). Thus, our goals of this symposium are to unify researchers from different fields, as well as provide opportunities to present novel findings, questions, and different perspectives in an effort to better understand the costs and benefits of deception.

D20SY19IT10:30R8

THE EVOLUTION OF IMPERFECT MIMICRY IN HOVER FLIES (DIPTERA: SYRPHIDAE)

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Although exquisite examples of the outcomes of natural selection are widely celebrated, it has long been recognized that nature's imperfections tell us more about the process of adaptation than its perfections. Here we consider the phenomenon of imperfect mimicry and the variety of evolutionary theories that have been postulated to understand it, concentrating on hover flies (Diptera: Syrphidae). First, we review what we know about the morphological, acoustic and behavioural similarity of hover fly mimics to their putative models (bees and wasps), asking whether mimicry is extended into the UV range of the reflectance spectrum, and whether the behavioural mimics tend to be good morphological mimics. Second, we take a phylogenetic approach to quantify how frequently evolutionary transitions between model types have evolved, and what features need to evolve to facilitate mimicry of a given model type. Taken together these empirical and comparative insights allow us to rule out some theories for imperfect mimicry and provide support for others, although there is much more to do to understand the maintenance of imperfect mimicry in this species rich group.

D20SY19IT11:18R8

IS VARIATION IN CHEMICAL DEFENCE PARASITISM ON A PUBLIC GOOD?

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An individual's chemical defences (toxins) contribute to a 'common good' by educating predators about the distastefulness of the population, hence deterring future attacks on the toxic individuals themselves and other individuals of similar appearance. Defensive chemicals are found in both simple and complex organisms including bacteria, fungi, animals and plants. Variation in defensive chemicals both within and among prey populations has been reported repeatedly in chemical ecology literature but it has received far too little attention from evolutionary ecologists. If some individuals lack defences ('automimics') they may exploit the common protection profiting from reduced attack rates but paying no individual cost of toxicity themselves. I will discuss whether variation in chemical defence and automimicry are examples of "environmental noise" or whether we need to seek evolutionary explanations for them. I will also discuss examples where understanding the dynamics of deception (e.g. Batesian mimicry) may have important consequences for successful conservation management.

D20SY19IT14:00R8

BROOD PARASITES AND MIMICRY: A SENSORY ECOLOGY APPROACH

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Cuckoos and other brood parasites are cheats – they lay their eggs in the nests of other birds, leaving all parental care to the hosts. A striking outcome of coevolution in many systems is egg and chick mimicry by parasites to deceive host parents into accepting young that they would otherwise reject. To understand interactions between host and parasite requires investigating the use of sensory information by both parties. I will discuss work my collaborators and I have done to understand several parasitic systems by considering the visual system of the receiver and the use of sensory information in decision-making (e.g. egg/chick rejection). I will show how mimicry of host eggs by parasites can be highly refined in terms of bird vision, and in turn how hosts use the most reliable information to discriminate between and identify their own and foreign eggs. I will then discuss how coevolution can drive different host species down alternative lines of defence, such as egg polymorphism, highly refined rejection behaviour, or chick rejection. Furthermore, I will discuss long-term data indicating how interactions between host and parasite can lead to rapid changes in egg phenotype over short timescales, and the nature of this change. Finally, I will discuss how parasites can exploit limitations in host sensory and cognitive systems to defeat host defences.

D20SY19RT14:48R8

RETALIATORY PARASITES MAKE AN OFFER A HOST SHOULD NOT REFUSE

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Mafia like behavior occurs not only in humans, but is also observed in animals. For example, experimental evidence suggests that avian hosts tend to accept a certain degree of parasitism in order to avoid retaliating punishment from the brood parasite. Herein, we model the interaction between hosts and parasites to understand under which conditions it will be beneficial for the host to accept parasitism. In our model, the host's behavior is plastic, and thus, its response depends on the previous interactions with the parasite. We find that such learned behavior in turn is crucial for the evolution of retaliating parasites. The abundance of this kind of mafia behavior oscillates in time and does not settle to an equilibrium. Our results suggest that retaliation is a mechanism for the parasite to evade specialization and to induce acceptance by the host.

D20SY19RT15:12R8

CONSEQUENCES OF AN ARMS RACE ON THE CUTICULAR HYDROCARBON PROFILES OF AN INSECT HOST AND ITS THREE BROOD PARASITES

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Parasites and their hosts have conflicting interests – to either successfully exploit the host or to defend against the parasite attack. This situation sets the board for an evolutionary arms race between both species. The species pair then follows a trajectory through repeated cycles of fine tuning of the parasite's attack strategies and evasive actions of the host. As a special case, brood parasites need to avoid detection by the host in order to neither be attacked while in the nest nor risk the nest to be abandoned by the host afterwards. Insect brood parasites may avoid olfactory detection by mimicking the host's cuticular hydrocarbon (CHC) profile. In this case, the arms race would lead to optimized chemical mimicry in the parasite. The host could e.g. change the CHC composition in order to escape a mimetic match. The most straightforward parasitic associations consist of one parasite and one host. More complex variations are possible but very rare: one parasite may use several hosts or several parasites may specialize in one single host. In this study, a solitary host and its three host-specific brood parasites serve as a model of such a multi enemy / single target system. We compare the CHC profiles and predict that a brood parasite whose intrusion is detectable by the host should develop chemical mimicry. The host in return should establish counterstrategies. Competition between parasites may fuel the perfection of mimicry or the development of completely new intrusion strategies. We find two chemotypes of the host that differ greatly in their CHC composition. Evolving a second CHC profile might be the outcome of escaping a parasite's mimicry. However, a second parasite species has evolved a close match of their CHC composition compared to the alternative chemotype. The third parasite produces its very own CHC bouquet – it has developed a new strategy for invading host nests and can no longer be fended off by the host.

D20SY19RT15:45R8

BATESIAN MIMICRY, MORPHOSPACE OCCUPANCY, AND THE SHAPING OF WARNING SIGNAL DIVERSITY IN BUTTERFLY COMMUNITIES

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Mimicry as a defensive strategy is one of the most compelling example of adaptation. Mimicry communities often involve numerous species, and both mutualistic (Müllerian) and deceptive (Batesian) mimics coexist. In deceptive mimicry, palatable prey mimic unprofitable species (e.g. chemically defended prey which predators avoid) with a negative impact on avoidance learning by predators. In mutualistic mimicry, appearances of defended prey converge on a similar warning signal, thereby reinforcing it and decreasing the per-capita cost of training predators. Theoretical and experimental studies on mimicry phenotypes abound, but studies empirically testing their predictions in real mimicry communities remain scarce. Indeed, the quantitative distribution of phenotypes within and among mimicry complexes is largely unknown, and how phenotypic variations are influenced by the type of mimicry, selection intensity, and/or phylogeny remains unaddressed. We first developed a novel framework that enables for the automatic and precise quantification and comparison of colour pattern. We then used this tool on over 2000 specimens, consisting of 130 butterfly species, collected from distinct Neotropical butterfly communities in the Peruvian Amazon. We quantified the distribution of phenotypes and their structure into a number of separate mimicry optima, using a morphological space encompassing the variation and frequencies of all coexisting colours patterns. We analysed this structure to extract the ecological and phylogenetic patterns underlying the coexistence of multiple mimicry groups within a given locality. We then demonstrated the influence of deceptive vs. mutualistic mimics on phenotypic variability around a mimicry optimum in order to address the effective impact of deception on mimicry.

D20SY19RT16:09R8

REPRODUCTIVE COMPETITION BETWEEN POLYGYNOUS PARASITE QUEENS IN ANT COLONIES

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One of the most remarkable and complex parasitic interactions is social parasitism, where a parasite exploits a complete societies, rather than an individual organism. By integrating into a society the parasite gains protection against predators and diseases, and can redirect resources from the host to increase its own fitness. Among the most specialized social parasites are the inquilines that exploit social insect colonies. Inquilines are usually close relatives of their host and so share ancestral characteristics (Emery's rule). They are dependent on being fully integrated into their host's colony throughout their lives in order to reproduce. Most inquiline ants have completely lost their sterile worker caste. An exception to this is Acromyrmex insinuator, a parasite of the fungus-growing ant Acromyrmex echinatior. Studies have shown that a threshold proportion of parasite workers in the colony are essential for parasite reproduction. Multiple invasions of parasite queens into host colonies suggest that each parasite queen may need to produce fewer parasite workers and that the reproductive phase can be achieved more quickly. Polygyny among parasite queens is expected to select for intraspecific hyperparasitism, where some queens might cheat by only producing sexual offspring, effectively parasitizing the worker force produced by other queens. We investigated hyperparasitism in A. insinuator by genotyping parasite offspring, workers and alates in polygynous nests to investigate any bias in the production of reproductive castes relative to workers. Hyperparasitism may provide a pathway towards speciation, and can shed light on the evolution of social parasites.

D20SY19RT16:33R8

SNEAKING BEHAVIOUR IN YOUNG MALES: ALTERNATIVE MALE MATING PHENOTYPES IN THE TWO-SPOTTED SPIDER MITE

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Sneaking behaviour as a strategy to compete for females has been observed in many taxa from insects to mammals. Often, the less competitive males who would lose contests over females against stronger males, display sneaking behavior in that they deceive their rivals by pretending to be female. To understand the evolution of observed sneaking behaviour, it is important to elucidate the mechanisms maintaining alternative mating strategies. Recently, we found three male mating phenotypes in a population of the two-spotted spider mite: territorial, sneaking and opportunistic. Opportunistic males wander around in search of females that are about to moult into the adult phase (teleiochrysalis). Territorial males spend much time in a mounted position on the dorsum of the teleiochrysalis females, and guard them by fighting intruding rival males. Sneaker males also spend much time in a mounted position, but they never show aggressive behaviour against intruders. Intruding males easily find and attack territorial males but fail to notice or even ignore sneaker males. Here, we focus on territorial and sneaker males in the two-spotted spider mite, and investigate which males display sneaking behaviour and how effective the sneaking behaviour is against territorial males. We show that young males display sneaking behaviour more often, and that young territorial males are three times more likely to lose their mounted position to old territorial males than young sneaker males. Old males are always territorial and rarely lose their position on the female dorsum to young males. Finally, young territorial males are observed to change their mating strategy to that of sneaker in the presence of old males. Our results suggest that old males are superior to young males in contests over females, and that in response to old males young males can phenotypically switch to sneaker behavior, thereby possibly improving their mating success.

D20SY19RT16:57R8

FLEXIBLE DECEPTIVE TACTICS OF THE FORK-TAILED DRONGO

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Animals commonly deceive each other, but just as in Aesop's fable 'The boy who cried wolf', deceptive signals cease to work when they are made too often. However, deceptive species might evade this frequency constraint by tactically varying their deceptive signal, thus maintaining their deception racket. My research investigates the behaviour of a South African bird, the Fork-tailed Drongo (Dicrurus adsimilis), which uses deceptive false alarm calls, including the mimicked alarms of other species, to scare host species and steal their food. Using a combination of observations and experiments undertaken on a wild population of individually recognisable drongos habituated to close observation, I show the benefits drongos gain from employing vocal mimicry to vary their false alarm calls. Drongos most frequently mimicked the alarm calls of a host species when using false alarms to steal food from that specific host. Furthermore, an experiment demonstrated that these mimetic alarm calls were more likely to deceive a host than the drongos own alarm calls. A second experiment showed that hosts reduced their response when the same false alarm call type was repeated, but increased their response again when the call type was changed. In natural conditions, drongos were more likely to change the type of alarm call they made when a previous false alarm call was unsuccessful and evidence indicates that this increased the likelihood drongos stole food. By tactically varying a deceptive signal, drongos exploit signals in the environment more likely to deceive a host, and potentially evade the frequency dependent constraints which typically restrict the pay-offs from deception. Results highlight that in communication systems, feedback from a receiver's previous response provides valuable information to signallers. Furthermore, they raise questions regarding the mechanisms that enable animals to produce apparently sophisticated communication behaviour.

POSTERS

D20SY19PS0220

CONTRASTING SHAPE, COLOR PLASTICITY AND HABITAT USE INDICATE MORPH-SPECIFIC ROLES IN A MARINE SHRIMP

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Color and shape polymorphisms are important traits of species, allowing a more generalist strategy and a better use of resources. An efficient occupation of space may lead to very abundant populations, triggering density-dependent processes and alternative mating systems. Hippolyte obliquimanus is a small gonochoristic shrimp species, and supposedly a generalist algal dweller in shallow waters. Yet, populations comprise two main morphotypes, homogeneous shrimps of variable color (H) and transparent individuals with disruptive stripes or bands (D). H color patterns tend to inhabit macroalgal substrates of matching background, while D individuals are evenly distributed between habitats. Unlike D shrimps, H morphs are capable of color change within a few days, but camouflage efficiency is habitat-dependent. Pink (P) animals collected in the red algal *Galaxaura* turned cryptic when supplied the brown weed Sargassum, but color change of greenish-brown (GB) shrimps from Sargassum did not fully conceal in *Galaxaura*. Homogeneous GB and disruptive morphs select Sargassum, while no preference was detected for homogeneous P individuals. Crypsis efficiency and habitat selection explain the much higher shrimp density in *Sarqassum* compared to *Galaxaura*. The overall population sex-ratio did not depart from the 1 : 1 ratio, but D individuals were mostly males and H shrimps chiefly females. These main morphs also differ in shape; D shrimps are more streamlined and H ones stouter, further suggesting enhanced mobility and substrate fidelity, respectively. Morph-specific functional roles would promote lower density and stable population dynamics at mixed algal beds, but higher density and a more fluctuating trend in monospecific *Sargassum* stands.

D20SY19PS0456

CATS DEVELOPED SPOTS AND STRIPES TO LURE RODENTS. A MULTIDISCIPLINARY FRAMEWORK FOR UNDERSTANDING THE FUNCTIONS OF THE COLOUR PATTERN ON CATS.

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To date the precise function of spots and stripes pattern on feline's fur remains a mystery. Predators use lures in the type and range of signals used by their prey to sense his environment in feeding activities and possible in other intra- and inter- species communication. The type of signals can be acoustic, visual, chemical, mechanical or other used by the prey. In our work we argue that predation by luring is a driving force to the formation of skin color patterns and behavior of cats. We propose a computational visual comparative model to demonstrate the function of the color pattern on feline's fur. We hypothesized that felines have developed a selective signaling, meaning that a signal pattern is intended to be detected by a specific receiver, but it would pass undetected for non-convenient receivers.

D20SY19PS0817

THE CICHLID AND REEF FISH VISUAL SYSTEM AS A MODEL FOR SPECIATION PROCESSES

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Freshwater cichlid and marine reef fish species represent spectacular products of adaptive radiations. Their amazing color diversities and well-adapted visual systems make them ideal candidates for a comparative study of molecular mechanisms involved in speciation based on the visual system. These will help to identify external factors and environmental conditions that shape color morphs and visual sensitivities. The major aims of this study are to compare (1) the coloration, (2) the molecular basis of the diversity of visual pigments, (3) visual pigment expression patterns, (4) transmission properties of the ocular media and (5) photic properties of habitats between cichlids and reef fish. Up to now, we sampled a representative set of shallow-water labrids (Labridae), damselfish (Pomacentridae) and cichlids (Cichlidae) in definite light habitats and studied their visual sensitivities composed of opsin gene variations and opsin gene expression patterns. Spectrophotometry was used to define the different light environments. To determine the molecular basis of visual pigments, we sequenced opsin genes using DNA and next generating sequencing. To quantify the relative amount of opsin gene expression we performed real time quantitative PCR.

19. Evolutionary Consequences of Deception

D20SY19PS1264

WARNING SIGNALS AND HANDICAP THEORY

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Aposematism has often been seen as a dynamical, transient phenomenon that can be destabilised by the appearance of undefended cheater prey (mimics). Recent theoretical work has made it clear that warning signals can also be evolutionarily stable and honest. Signalling theory predicts that an honest signalling system can contain a limited number of cheats without being destabilised as long as the system remains 'honest on average'. Somewhat analogous to this, a game theoretical analysis of three different mechanisms for honest warning signalling predicted an overall positive relationship between conspicuousness and defence across prey, given that signals were honest on average. Here we extend this analysis further and identify mechanisms for honest warning signalling that do not give rise to a positive relation between conspicuousness and defence. Some complicating factors include tradeoffs between warning signals and life-history traits, and non-monotonic relations between prey aversion and toxicity that may arise when predators can taste-reject prey.

19. Evolutionary Consequences of Deception

D20SY19PS1442

MIMICRY EVOLUTION: IT'S IN THE EYE OF THE BEHOLDER

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During mimicry evolution the perceived mimic-model similarity increases and the evolutionary direction could be strongly influenced by predators judgement of similarity. Mimicry evolution is thus more likely to commence in traits that predators use prior to others to categorise prey. If they use such feature-based categorisation, a similarity with the model in such a feature should lead to increased survival. I tested the idea by studying if birds attend to specific features of prey appearance when they learn to discriminate and generalise between them. I used wild Blue Tits as predators and tested the colour, pattern and shape dimension of artificial prey. The birds first learned to avoid a specific model prey appearance and then performed a generalisation test with new mimics that shared one dimension with the model. I found that colour mimics were strongly avoided whereas pattern and shape mimics were attacked. This shows that the birds primarily attended to a single feature, colour, and thus generalised between models and colour mimics. In a separate experiment I found that the birds learned the colour dimension significantly quicker than pattern and shape, showing that colour is a significantly more salient trait to them in terms of learning and categorisation.

Symposium

20. Mechanisms of Trade-offs

23 August



Program

Friday 23 August

Session(s): 10, 11, 12, 13

Organisers: Bas Zwaan and Thomas Flatt

Invited speakers: Stephen Stearns

Description:

Trade-offs are key in evolutionary theory: traits can only evolve in the direction of increased fitness at the expense of loss of fitness through other traits. Because organisms are universally restricted in resource acquisition, trade-offs have been explained by differential resource allocation. Recent molecular data has challenged the validity of this view. Unfortunately, up to date, trade-offs have either mostly been approached from a purely phenotypic perspective without much attention to the underlying physiological mechanisms, or conclusions have been drawn about trade-offs from molecular studies without considering the functional phenotype. Here, we aim to bridge these views by linking our current knowledge of the molecular and physiological pathways with that about quantitative genetic and phenotypic correlations between traits.

D23SY20IT10:30R4

COMPARING PHENOTYPIC, QUANTITATIVE GENETIC, AND GENOMIC APPROACHES TO MEASURING TRADEOFFS IN A CONTEMPORARY HUMAN POPULATION

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In women born before 1940 in Framingham, Massachusetts, there was a significant negative correlation between number of children ever born and lifespan. Each additional child was associated with a reduction of about one year of life. Analysis of the 1500+ pedigrees containing 15,000+ people with an animal model that partially controlled for cultural and environmental effects yielded a large, significant, negative genetic correlation between children ever born and lifespan. A genome wide association study that looked for genes that alter the slope of the relationship between those two traits discovered several genes, some of which have previously been identified as involved in cancer. One, EOMES, has the sort of function one would expect of a gene that influences a tradeoff: its product affects many different processes. Statistical models with and without education as a covariate yielded results consistent with the idea that education is a cultural mimic of antagonistic pleiotropy: increases in level of education are associated with fewer children and longer life.

D23SY20RT11:18R4

EVOLUTION OF FOOD PREFERENCES DRIVES PLEIOTROPIC FITNESS TRADE-OFFS IN BACTERIA

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Bacteria face trade-offs between life history strategies, among which the choice of a generalist or specialist resource use strategy has important effects on fitness. Bacteria bypass this problem by prioritizing nutrient utilisation. They highly specialise on preferred nutrients when available, and invest in a generalist way of life when only second-choice nutrients are left. Here we show that bacteria rapidly evolve new food preferences as an adaptation to a new substrate. We follow the expression of the small regulatory RNA crcZ in Pseudomonas fluorescens which is expressed in absence of preferred resources. It activates the translation of genes involved in alternative resource use. We show that the substrates inhibiting crcZ levels rapidly change when bacteria are confronted with second-choice subtrates, indicating that bacteria evolve new preferences. We further show that these altered food preferences affect the use of complex substrates by bacteria as well as their coexistence with competitors. Evolution of food preferences by bacteria appear therefore as efficient strategy to adapt to new conditions by readjusting the regulation of genes linked to substrate uptake and catabolism.

D23SY20RT11:42R4

MICROBIOME MEDIATES OXIDATIVE COSTS OF REPRODUCTION

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Parasites exert important selective pressures on host life-history traits. By affecting allocation tradeoffs between reproduction and self-maintenance, they have major consequences for host reproductive success and survival. Microbiome is a major part of earth biomass, including pathogenic and commensal microorganisms. To date experimental studies that examine how the microbiome shapes host life-history traits are still lacking. Here we modified nest microbial communities of wild breeding great tits (*Parus major*) to test whether the microbiome mediates the oxidative cost of reproduction. We found that microbiome affected the relationship between fledgling number and adult oxidative damage. Adults raising a large number of nestlings show higher oxidative damage in control and high bacterial density treatments, whereas this trade-off was absent when decreasing bacterial densities. This study provides the first experimental evidence for a role of environmental microbiome in mediating the oxidative costs of reproduction. Our results show that the microbiome may constitutes a fundamental factor shaping animal life-history traits that need to be considered in future studies.

D23SY20RT14:00R4

HSP83 AND TOTC ARE REQUIRED FOR PATHOGEN-INDUCED HORMESIS IN THE FRUIT FLY

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Hormesis contradicts the fundamental evolutionary concept that organisms can't have it all. Many studies have found that treatments, such as limited stress and diet restriction, can enhance Darwinian fitness without obvious costs. This is a problem for our understanding of trade-offs as it suggests both that organismal fitness is sub-optimal, and that organisms can avoid trade-offs between life history traits. However, few studies have considered the possibility that hormesis is driven by a trade-off between Darwinian fitness and pathogen resistance. Here we show that topical treatment with an inactive fungus increases the survival and fecundity of various strains of the fruit fly. Using mutant strains and the Gal4::UAS knockdown system, we show that the heat shock protein Hsp83, and the generalist stress gene TotC, are required for this response. Preliminary evidence suggests that this hormetic response depends both on the temperature and diet of the host. These results are important as they identify the genetic basis of a novel hormetic response and provide a potential explanation for phenomena which appear to defy the constraints apparent in life history trade-offs: that hormetic responses are only beneficial under a narrow range of ecological conditions. Thus, our results are consistent with the hypothesis that hormesis also involves trade-offs.

D23SY20RT14:24R4

TRADE-OFFS BETWEEN PERSONAL IMMUNITY AND REPRODUCTION IN THE BURYING BEETLE (*N. VESPILLOIDES*)

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Immunity and reproduction are both costly, but how do organisms balance these costs when called upon to carry out both at the same time? We tested this with the burying beetle, *Nicrophorus vespilloides*. This species exhibits costly bi-parental care and its immune system can be readily quantified. The trade-off between self-maintenance and reproductive investment is likely to be most visible in those species that invest extensively in parental care, making *N. vespilloides* a valuable study species. Immune function was measured during the breeding bout in both immune-challenged and control individuals. Breeding success in the current bout and across each individual's lifetime was then measured to assess the potential costs associated both with maintaining the immune system in a state of readiness and deploying an immune response. Deciphering the mechanistic basis of trade-offs is important. Hormones are increasingly being invoked as a potential mechanism by which life-history trade-offs are controlled. I will discuss the potential of Juvenile Hormone as the candidate for this role in *N. vespilloides*.

D23SY20RT14:48R4

HOW DO AGGRESSIVE FISH WIN FIGHTS? AN INSIGHT FROM TRANSCRIPTOMICS

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Aggressive behaviour can be beneficial, for example by allowing individuals to acquire territories and other limited resources. In juvenile salmonids, aggressive behaviour carries energetic costs, such that aggressive individuals tend to grow at slower rates than less aggressive individuals. To understand the molecular mechanisms underlying aggressiveness, rainbow trout Oncorhynchus mykiss were assessed for aggressive behaviour by dyadic contests. Aggressive fish were those that performed more aggressive acts during a contest and less aggressive performed fewer. Using next generation sequencing, the transcriptomes of each behavioural type were characterised. Genes previously implicated in aggressive behaviour in vertebrates, such as POMC, were differentially expressed. In addition, a high proportion of differentially expressed genes were associated with metabolism, where aggressive animals upregulated more genes associated with oxidative stress and with oxygen binding, whereas less aggressive trout expressed genes associated with increased respiration. These findings indicate the mechanisms by which aggressive and less aggressive rainbow trout differentially allocate resources during an interaction and may demonstrate an underlying propensity for competitive ability. More broadly, these results show the utility of using next generation sequencing to understand the networks of genes involved in behavioural trade-offs.

D23SY20RT15:12R4

LIFE-HISTORY CONSTRAINTS ARE EXPOSED WHEN GENETICALLY-MODIFIED MICE COMPETE WITH CON-SPECIFICS

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An organism's investment in life history traits such as growth, reproduction and lifespan will be constrained by a variety of physiological factors. With regards to constraints on lifespan, genetic modification of gene expression has been successfully used to uncover some of the molecular pathways that influence how long an organism lives. These techniques have the potential to reveal constraints on other life history traits as I will demonstrate in this talk. We used a genetically-modified strain of mouse to test how various components of life history are constrained by oxidative stress, a physiological condition also implicated in ageing. Mice that did not express a key antioxidant enzyme used to protect against oxidative stress, copper-zinc superoxide dismutase (Sod1), showed altered investment in behavioural, morphological and molecular aspects of reproduction and sexual signalling. These effects were also found to be more prominent when animals were maintained in a competitive environment. We then used phenotypic manipulations to increase investment in various reproductive traits in both genetically-modified and wild-type animals. This allowed us to determine how oxidative stress, the outcome of this gene knockout, influences an organism's ability to increase metabolic rate, adjust mitochondrial function and limit further oxidative damage when investing in demanding periods of reproductive effort. Our results reveal that oxidative stress is one aspect of physiology that can directly reduce investment in reproduction. The examination of genetically modified animals in more ecologically-relevant conditions offers exciting opportunities to uncover the mechanistic constraints on life history evolution.

D23SY20RT15:45R4

PHENOTYPIC ENGINEERING OF TESTICULAR AND OVARIAN FUNCTION REVEALS A SEX ALLOCATION TRADE-OFF IN A SIMULTANEOUSLY HERMAPHRODITIC FLATWORM

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A resource allocation trade-off between investment towards the male and the female function is a fundamental assumption of sex allocation theory for simultaneous hermaphrodites, but there is currently limited empirical support for it, especially among animals. Here we experimentally manipulated testicular and ovarian function in the free-living flatworm, Macrostomum lignano, and subsequently tested for the occurrence of the sex allocation trade-off. Specifically, we used RNA interference—by soaking worms in a solution of double-stranded RNA—to knock down the expression of specific candidate genes, which we identified in two ways. First, we screened 11 gonad-specific candidate genes obtained from a published microarray study in the distantly related planarian flatworm, Schmidtea mediterranea. Seven of these candidate genes had testis-specific expression in *M. lignano*, and the knockdown of five of these had evident effects on testicular function. The most prevalent effect was a disruption of sperm production, which for one candidate gene was accompanied by an increase in ovary size. Second, using an RNA-Seq screen in *M. lignano* we identified a candidate gene with ovaryspecific expression, knockdown of which prevented the formation of yolk in developing oocytes and also resulted in larger testes (and a trend for a higher sperm production rate). Both of our approaches thus yielded candidate genes whose knockdown uncovered sex allocation trade-offs (notably via manipulation of both the male and the female function). This, to our knowledge, is the first study to use phenotypic engineering of testicular and ovarian function to empirically test the trade-off assumption of sex allocation theory. Furthermore, our results suggest that disrupting the function of one gonad does not necessarily lead to a trade-off, possibly because some types of disruption do not liberate resources that become available to the opposite function.

D23SY20RT16:09R4

EXPERIMENTAL INSIGHT INTO CONDITION-DEPENDENT TRAIT EXPRESSION: NEAR-ORTHOGONAL EFFECTS OF ENVIRONMENTAL VS. GENETIC MANIPULATIONS IN DROSOPHILA MELANOGASTER

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Differences in condition among individuals arise from both genetic and environmental variance in their acquisition and/or assimilation of resources. Such variation in condition can weaken or otherwise obscure the physiological trade-offs among competing life history traits that are thought to necessarily arise from the allocation of finite resources. The nature and extent of among-individual variation in condition will therefore determine whether life history traits co-vary positively or negatively and is therefore central to our understanding of life history trade-offs. Our understanding of conditiondependent trait expression derives almost exclusively from environmental manipulations, yet it is the underlying genetic basis of condition, and the pleiotropy this generates among life history traits, that may have fundamental evolutionary consequences. A comprehensive understanding of life history traits, their trade-offs, and their evolution, will therefore require a detailed knowledge of the impacts on trait expression of both environmental and genetic variation in condition. Here we use a two-way factorial design to provide some of the first experimental data comparing the effects of diet and mutation-accumulation (i.e. environmental and genetic) manipulations of condition on a suite of sexual displays (epicuticular pheromones) and morphological traits in Drosophila melanogaster. Our results reveal that condition is multi-dimensional, with the effect of the environmental manipulation being almost orthogonal to that of the genetic manipulation for both sets of traits in both sexes. Only body size showed concordant effects of diet and mutation accumulation. This suggests that environmental manipulations alone may provide misleading insight into condition-dependence and its effects on the expression, evolution, and trade-offs among competing traits.

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TRANSCRIPTOMICS OF A LIFE HISTORY TRADE-OFF IN THE WING POLYMORPHIC CRICKET (GRYLLUS FIRMUS)

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The sand cricket *Gryllus firmus* is a model species in the study of the trade-off between (flight) dispersal and fecundity associated with wing-polymorphism in insects. *G. firmus* females exist as either a flight capable, long-winged (LW(f)) morph that delays egg production or as a flight-incapable, shortwinged (SW) morph with substantially elevated egg production. In addition, some adult LW(f) individuals partially degenerate their flight musculature, thus giving rise to a third morph (LW(h)), that exhibits the substantially increased egg production seen in the SW morph. Here we present findings of a transcriptomics study that examined molecular mechanisms underlying plasticity in flight muscle and fat body function among the *G. firmus* morphs. Expression of genes encoding proteasome components was significantly enhanced in LW(h) flight muscle, however we observed a striking, nearly complete absence of differences in flight muscle sarcomere gene expression between LW(f) and LW(h) morphs. This indicates that flight muscle histolysis in the LW(h) morph is a highly controlled and selective process, and that maintenance of non-histolyzed fibers in LW(h) flight muscle is controlled through gene expression patterns similar to that in the LW(f) morph. In addition, LW(h) morph flight muscle displayed a significant reduction in expression of genes encoding aspects of mitochondrial structure and metabolism, supportive of earlier enzymological and metabolic rate work in this species. Genes involved in triglyceride biosynthesis were significantly upregulated in LW(f) fat body tissue, while SW fat body exhibited substantially elevated expression of an insulin-like peptide known to regulated egg production in locusts, as well as a number of lectin-related proteins that function in immunity. We will discuss our findings with respect to the physiological and molecular mechanisms underlying the flightfecundity trade-off in *G. firmus* and other insect species.

D23SY20RT16:57R4

LINKING PROTEIN STRUCTURE AND TRADE-OFF BETWEEN LIFE-HISTORY TRAITS

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Metabolic pathways implicated in external resource consumption are expected to have an essential role in the evolution of life-history traits. Two extreme life-history trait strategies related to different resource exploitation strategies have been described in the yeast, Saccharomyces cerevisiae : The "grasshoppers" have a high specific glucose consumption rate in fermentation, a large cell size but a low reproduction rate and carrying capacity, while the "ants" have a low specific glucose consumption rate, a small cell size and a high reproduction rate and carrying capacity (Spor et al. 2008, 2009). We have explored the genetic and metabolic bases of « ant » and « grasshopper » life-history strategies. Using mutant analysis, we have shown that variation of expression of glycolytic encoding genes allow moving on the continum between these two extreme life-history strategies (Wang et al. 2011). Using proteomics appoaches we have revealed that not only glycolytic gene expression is involved in the control of yeast life-history strategies but also post-traductionnel modification of specific glycolytic enzymes (Albertin et al. 2013). Finally using experimental evolution, we have found that the evolution towards « ant » or « grashopper » strategies can be explained partly by mutations in a highly pleiotropic gene involved in a hub of more than 200 protein interaction network. This gene is recurrently mutated in the laboratory evolution. The location of the mutations in the gene, and thus in the protein, changes the effect of the mutation on several traits. Altogether, our data highlight the role of both variation in gene expression and also protein structure in the trade-off between life-history traits.

D23SY20RT17:45R4

TESTS OF LIFE-HISTORY THEORY UNDER SELF- AND CROSS-FERTILIZATION IN CAENORHABDITIS ELEGANS

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In organisms that are able to reproduce by self-fertilization (selfing) and by cross-fertilization (outcrossing), a fitness trade-off between both reproduction modes is necessary for them to be maintained during evolution. To assess if the maintenance of selfing and outcrossing at intermediate frequencies is in part due to the evolution of resource allocation towards the male or the female functions in hermaphrodites, we performed experimental evolution in *Caenorhabditis elegans*, an androdioecious nematode in which hermaphrodites are self-fertile but can also be cross-fertilized by males. For this, we characterized the reproductive schedules and the longevity of hermaphrodites under selfing or outcrossing with males that did not have the opportunity to evolve with them. Hermaphrodite life-history was compared to that of females, which evolved under similar environmental conditions but with obligate outcrossing. After 100 generations of evolution, the reproductive investment of hermaphrodites when selfed was shifted towards earlier ages, as expected with selection for early reproduction, together with an increase in mean longevity. When hermaphrodites were outcrossed no such changes in reproductive investment or longevity were observed. Like selfed hermaphrodites however, females from the obligate outcrossing populations showed increased earlier reproductive investment and increased longevity. These findings suggest that the fitness trade-off between selfing and outcrossing in experimental *C. elegans* populations is due to the evolution of resource allocation towards male functions in hermaphrodites.

D23SY20RT18:09R4

THE REPRODUCTION-SURVIVAL TRADE-OFF AT THE TRANSCRIPTIONAL LEVEL

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To reproduce, organisms typically compromise their own immediate and future survival. Trade-offs are pervasive and form an important component of many biological theories. Despite the considerable attention dedicated to trade-offs over last years and sufficient genomic tools, it remains unclear how the patterns in gene expression translate to life-history trade-offs observed at the phenotypic level, and to what extent the knowledge at the molecular and theoretical levels can be integrated. Here, we report that the genes functionally linked to reproduction and survival in Drosophila melanogaster show negatively correlated expression patterns, mirroring the reproduction-survival trade-offs observed at the phenotypic level. We further show that the expression trade-off mediates responses to food and age in accordance with life history theory. This includes a parallel shift of the trade-off in females on different diets complying with the acquisition-allocation theory, and a trade-off release in males on high food levels following the notion of lower reproductive investments in males. Taken together, our findings (i) indicate that life history trade-offs can be interpreted as conflicts over gene expression, (ii) extend the interpretations of basic concepts in biology to the transcriptional level, and, (iii), suggest mechanisms of how these trade-offs are regulated. Up to date, trade-offs have either mostly been approached from a purely phenotypic perspective without much attention to the underlying mechanisms, or conclusions have been drawn about trade-offs from molecular studies without considering the functional phenotype. Our study clearly bridges these views and provides a novel molecular perspective for interpretation and testing of classical biological theories.

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TESTOSTERONE MEDIATION OF A TRADE-OFF BETWEEN OFFSPRING CARE AND REPRODUCTION IN THE COOPERATIVELY-BREEDING BANDED MONGOOSE (MUNGOS MUNGO).

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Investment in offspring care commonly leads to a reduction in survival and fecundity, generating a trade-off between care of current young and future reproduction. Theory suggests that variation in this trade-off may explain the conspicuous individual differences in helping effort observed in cooperative breeders. In non-social species, testosterone is often found to mediate the trade-off between offspring care and reproduction. However, little is known about the mechanism mediating the trade-off between helping and future reproduction in cooperatively-breeding vertebrates. Here, we investigate testosterone as a candidate mechanism mediating trade-offs between cooperative offspring care and reproduction in the banded mongoose (Mungos mungo). Periods of high investment in offspring care are preceded by low faecal testosterone metabolite (fT) concentrations, suggesting that testosterone may inhibit offspring care in this species. During group oestrous, high ranking individuals with access to mates show elevated fT concentrations and a concurrent decrease in offspring care investment. However, outside of group oestrous, when there are no available mating opportunities, we find no correlations between individual rank and fT concentrations or investment in offspring care. These results provide evidence for testosterone mediation of the trade-off between offspring care and current reproduction in the banded mongoose, similar to that seen in non-social species. However, the mechanism mediating the trade-off between offspring care and future reproduction remains unknown. Together, these results highlight understanding the role of physiological mechanisms in mediating behavioural trade-offs as a key area of study to explain within-group variation in cooperative investment.

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GENETIC TRADE-OFFS AND THE EVOLUTION OF HUMAN LIFE-HISTORIES

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Understanding life-history trade-offs is fundamental to explaining the diversity of life-history strategies in nature, and determining the genetic basis of trade-offs can identify how evolutionary constraint maintains life-history variation. Humans have evolved an unusual life-history compared to other primates, characterized by the menopause and long female post-reproductive lifespan (PRL). These have been hypothesized to evolve to enable (i) reduced reproduction when the costs of reproducing exceed the benefits, and (ii) enhanced grandchild survival. Previous tests of these hypotheses have examined phenotypic correlations between female reproductive rate and (i) PRL and (ii) offspring survival. However, environmental effects can mask genetic associations, and the direction and magnitude of these genetic correlations must be examined to determine the evolutionary potential of such traits. Using genealogical data from preindustrial Finnish church records for eight populations, we applied a multivariate quantitative genetic framework to examine the genetic basis of female reproductive rate, measured by inter-birth interval (IBI). We examined how additive genetic effects on IBI changed with age, and how age-specific genetic effects varied across environmental conditions. We determined the genetic trade-offs between IBI and both PRL and offspring survival, and how these trade-offs varied across ages and environments. Pilot analyses on four populations show a genetic basis to all traits, and suggest that genetic trade-offs between IBI and the other traits increased with age in poor environmental conditions, but were weak and age-independent in good conditions. IBI and PRL were positively related to lifetime fitness, suggesting that genetic trade-offs act as an evolutionary constraint. Our results will reveal new insight into human life-history evolution and generally highlight the fact that genetic correlations between traits may be age- and environment-dependent.

POSTERS

MASKING SICKNESS IN PUBLIC RESULTS IN COSTS FOR THE MAGNITUDE OF THE IMMUNE RESPONSE

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Animals suffering from an infection undergo behavioural and physiological alterations that potentiate the immune system in fighting the pathogen. "Sickness behaviour" is the name given to the behavioural component of this response and consists of a generalized reduction in physical activity. Adopting this state of low activity is thought to free resources for mounting an immune response. However, an increasing number of reports indicate that these sickness behaviours are plastic and can be overcome in the face of conflicting adaptive opportunities, such as mating or parental duties. Here, we tested whether this plasticity imposes costs on the magnitude of the immune response. Using captive colonies of zebra finches (*Taeniopygia guttata*), we developed a miniaturized device that could be attached to the birds and quantify activity remotely. We housed birds either in isolation or in their colony and induced sickness behaviours by means of a lipopolysaccharide (LPS) injection. We found that birds housed in a group and injected with LPS exhibit reduced symptoms of sickness behaviours as compared to their isolated counterparts. This reduction of sickness behaviours was associated with reduced immune response. The findings here indicate that these animals face potential costs when reducing expression of sickness behaviours. The importance of the affected components of immunity for fighting a particular infection will determine the extent of these costs.

AGEING AS A TRADE-OFF MECHANISM: WHAT TELOMERES ACTUALLY TELL US?

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Telomeres are chromosome ends that protect the cell's genomic integrity. Telomeres are lost at a variable rate as a result of the imbalance between pro- (oxidative stress) and anti-erosion processes (telomerase), and once critically shortened, telomeres can trigger cell death (apoptosis). Consequently telomere function has been one of the main cell mechanisms studied in ageing biology. Recently, it has been proposed that telomeres might be critical for our understanding of life-history trade-offs in general. Telomere length is heritable and at the organism level telomere length has been shown to correlate with individual longevity, first in humans but also in wild animals, appearing as a strong proxy of individual survival. Furthermore, telomere length was recently proposed to reflect individual phenotypic quality in terms of high rates of reproduction. All these studies suggest that telomere dynamics may be one of the key genetic determinants explaining the individual variability in ageing and fitness. However, the nature of the determinants leading to changes in telomere length early in life, during development, and of the mechanisms that link telomere length to fitness remain unknown. In this talk, I will present some of the data collected by our group which is studying how telomere length may be implicated in life-history trade-offs. For this I will show how an experimental manipulation of the cost of reproduction in zebra finch may affect telomere loss of breeders, but also how this brood size manipulation has affected telomere length in chicks. In a second step, I will show how telomere lengths are varying in early life in two wild species, great and coal tits, and how these variations may be link to local adaptation to favour chick growth and survival. Finally, as a short perspective, results of a pilot study on telomerase activity will propose a hypothetical pathway by which telomere biology may influence individual fitness.

MODELING THE RELATIONSHIP BETWEEN AGE SPECIFIC FECUNDITY AND LIFESPAN IN A COHORT OF FRUIT FLIES

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Populations of laboratory animals that are selected for increased lifespan often show correlated negative responses in early fecundity. However in some cases late fecundity, or total lifetime fecundity, is higher in the populations selected for increase lifespan. By some, this has been interpreted as a falsification of the disposable soma theory. According to the Y–model, in which the effects of variation in allocation and acquisition on life histories are studied, an alternative is suggested. A negative relationship between lifespan and reproduction can be viewed as variation in allocation, whereas a positive relationship is the result of variation in acquisition. We have analyzed age specific fecundity and lifespan in a cohort of Drosophila melanogaster flies which were individually housed. Early fecundity related negatively with lifespan, while late fecundity related positively with lifespan in the same cohort. We show that a model which incorporates the ideas of the Y-model, disposable soma theory and a decrease in physiological performance when age increases, can explain how the relationship between fecundity and lifespan changes with age. Furthermore, we modeled different environments in which there is variation in extrinsic mortality rates. In high mortality environments there was selection for high early fecundity, low late fecundity and low lifespans, whereas the opposite was true for low mortality environments. Our laboratory population of *D. melanogaster* was founded from a combination of flies from different areas in Europe and therefore is heterogeneous in genotype. We conclude that the differences in life history strategies found in one cohort of laboratory flies are the result of a mosaic of selection on the relationship between fecundity and lifespan over age.

INDIVIDUAL HETEROGENEITY AND A TRADE-OFF BETWEEN TWO COMPETING LIFE-HISTORY TRAITS WITHIN SPECIES

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A trade-off between two (or more) competing traits is widely observed at the inter-specific level. Therefore, it is often assumed that the same trade-off will be readily observable at the intra-specific level. However, this is often not the case because differences in individual quality or individual heterogeneity within a species can override a negative correlation between two competing traits (i.e. a trade-off). That is, individual heterogeneity can create a positive relationship between two competing traits within a species, a completely opposite pattern expected from life history theory. Although a classical theoretical model predicted this positive relationship at the intra-specific level, the generality of the effect due to individual heterogeneity is not systematically investigated before. Here, we use meta-analytic techniques to investigate two within-species relationships between maternal quality (size) and offspring size, and between maternal quality and offspring number. We show the pervasiveness of individual heterogeneity effects on the relationship between the two competing traits (i.e. offspring size and number) across taxa. In the light of such results, we discuss difficulties in studying trade-offs within a species, and argue the importance of comparative approach to study the mechanisms of trade-offs.

COMPARISON OF OXIDATIVE STRESS RELATED ENZYME ACTIVITIES IN TWO ATLANTIC SALMON (SALMO SALAR L) POPULATIONS: INSIGHT TO THE TRADE-OFFS BETWEEN PHYSIOLOGY AND SURVIVAL DURING EARLY LIFE STAGES

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Stressful conditions experienced by individuals during their early development might have long-term consequences on various fitness-related traits and ultimately their survival. Moreover, oxidative stress (OS) has been identified as a significant physiological constraint affecting trade-offs between reproduction, growth and survival. Several studies have also suggested the level of OS to be heritable, yet very little is known about the relationships between OS, fitness and survival of natural populations in wild habitats. Here, we measured the activity of three OS related enzymes: glutathione reductase (GR), glutathione-S-transferase (GST) and superoxide dismutase (SOD) during early life stages in two Atlantic salmon populations (in total 52 half-sib families) and evaluated the importance of OS related enzyme activities to survival and growth of juvenile fish in multiple natural environments. We identified significant differences in measured enzyme activities, growth rates and survival both within and between populations. In addition, we found that significant heritability exists for GR in Narva (h2=0,26; 95%CI 0,09-0,55) and GST activities in Kunda (h2=0,28; 95%CI 0,13-0,61) and in Narva (h2=0,75; 95%CI 0,51-0,91) but not for SOD. We conclude that the inter-population differences in OS during early life stages may explain significant proportion of variation in fitness and survival in wild.

EXTRINSIC MORTALITY AND LIFE SPAN IN BIRDS AND MAMMALS

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Life history theory predicts that species experiencing high extrinsic mortality will, on average, evolve shorter life spans than species that experience low extrinsic mortality. Perhaps the most striking evidence for a link between extrinsic mortality and life span comes from comparing bats with nonvolant mammals. On average, bats live 3.5 times longer than similarly sized placental mammals, presumably because they can fly and thus more easily escape predation. However, it is hard to test this as bats are the only volant mammals. In addition, flight is not the only way of reducing the risk of extrinsic mortality; other ecological factors may also be important such as activity patterns, foraging environment, and group size. A solution to this is to investigate correlates of longevity across all endotherms. Here we aim to determine which ecological variables related to rates of extrinsic mortality (specifically flight capability, activity pattern, foraging environment and fossoriality), are most important for predicting longevity in endotherms. We split species into volant (most birds and bats), and non-volant subgroups, because we expect sources of extrinsic mortality, and thus correlates of longevity, to differ in these two groups. We test our predictions on a large dataset (N = 1350 species) and use state-of-the-art phylogenetic comparative analyses (including a recently published bird phylogeny). Our results confirm that volant species live longer than non-volant species after controlling for body mass, but also show that the effects of body mass on longevity in these groups differ substantially. We also find that for volant species, life span is determined by when a species is active, whereas for non-volant species, life span is determined by where a species is active. This is the first time such an analysis has been conducted across such a broad range and number of species, and suggests that with respect to life span, bats should be considered birds.

OPTIMAL FORAGING OF LITTLE EGRETS AND THEIR PREY IN A FORAGING GAME IN A PATCHY ENVIRONMENT

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To avoid extinction, native species will rely increasingly on their ability to adapt to human-dominated habitats. Will the behaviors that evolved in natural circumstances help species make the transition? Or will they be maladaptive? A laboratory theater containing three fish pools with little egrets, Egretta garzetta (the predator), and common goldfish, Carassius auratus (its prey) help to pose this question usefully. The two species were playing a behavioral game in which their best tactics depended on how the other species was behaving. We tested the hypotheses that the egrets maximize their total capture success by responding to the fish's anti-predatory behavior and that the behaviors of both players respond adaptively to the density distribution of fish among the pools. Within each pool, fish could move between a safe, covered microhabitat and a risky, open microhabitat. Only the risky habitat had food, so fish were trading off food and safety by allocating the time spent in the two habitats. Egrets spent more total time in pools with more fish and returned to them sooner. Egrets maximized the number of fish they captured by following the matching rule of the ideal free distribution. Fish finetuned their behavior by reducing their use of the risky habitat as the egrets increased the frequency of their visits. It seems that egrets and fish adjust adaptively their behaviors by responding and counterresponding behaviorally to each other and to the experimental conditions in a way that tends to maximize capture success for the egret and tends to minimize the probability of being eaten for the fish. Apparently, the tactical rules that these species evolved in natural habitats prepared them also to deal effectively with this anthropogenic one.

INBREEDING DEPENDENT TRADE-OFFS IN THE ANT FORMICA EXSECTA

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Trade-offs between different defence mechanisms and life-history traits are known to exist. Especially interesting are the ones involving immune defences, as surviving infections is paramount for any organism. The capability to respond to stressors depends on the phenotypic plasticity of an organism. It is assumed that inbreeding decreases the adaptability of organisms to environmental stresses (e.g. dealing with pathogens, oxidative stress). Recent evidence from many insects highlights the importance of Reactive Oxygen Species (ROS) in insect immunity by regulating potential pathogens and keeping commensal gut flora under control. Although the release of these free radicals is an extremely effective defence against intruders, it is also harmful to the organism itself, as it causes oxidative stress. This is a situation where cellular production of ROS overwhelms its antioxidant capacity, leading to damage of various macromolecules (e.g. lipids, proteins, DNA). Dealing with this self-inflicted injury is essential, but requires extra resources for anti-oxidant production. In a multi-infection scenario, the capability of an organism to deal with multiple stressors may give a crucial advantage in comparison to less adaptive phenotypes. Here, we study how ants with different inbreeding levels cope with bacterial and fungal parasites, being previously exposed to oxidative stress via feeding with ROS. Dietary intake of extra ROS may lead to better survival against gut penetrating entomopathogenic bacteria, but may at the same time leave fewer resources to fight against fungal pathogens. We use oral infection with the pathogenic bacterium *Serratia marcescens*, and the exposure to the generalist fungal pathogen Metarhizium brunneum to study the trade-offs between ROS responses and the immune response in workers of F. exsecta. Comparing inbred colonies and outbred colonies reveals differences in the expression of various antioxidant and immunity related responses.

LIVE SMART, DIE YOUNG – SEX DIFFERENCES IN COGNITIVE AGEING IN THE DIOECIOUS NEMATODE, CAENORHABDITIS REMANEI

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Learning and memory are costly and are known to trade-off with reproduction and longevity in invertebrates. The most striking example is a symmetrical evolutionary trade-off between learning ability and longevity in Drosophila melanogaster. It has been found that populations selected for improved learning suffered a considerable reduction in longevity, while populations selected for increased longevity suffered a reduction in learning ability. Since males and females often resolve their survival-reproduction trade-offs differently, it is likely that the trade-off between learning ability and longevity will also be resolved differently between the sexes. Applying the logic of symmetrical evolutionary trade-off, increased investment in learning ability early in life should result in decreased longevity in a given sex. On the other hand, the longer-lived sex would experience stronger selection late in life, such that it should maintain its level of performance for longer than the shorter-lived sex. Here, we used a positive associative learning paradigm in a dioecious nematode worm *Caenorhabditis* remanei, which exhibits strong sexual dimorphism in lifespan, to test whether sexes differ in agerelated learning performance. Specifically, we tested whether the shorter-lived sex (females in C. *remanei*) will exhibit increased learning ability early in life and whether the longer-lived sex will maintain its level of performance for the longer period of time. Indeed, we found that young females outperformed young males in making a novel association between an odour (butanone) and food (bacteria). Moreover, while learning ability declined rapidly with age in females, males maintained higher learning ability until middle age. These results not only demonstrate sexual dimorphism in agerelated learning ability but also suggest that it is in line with sexual dimorphism in life-history.

ON THE EVOLUTION OF LIFE HISTORY TRADE-OFFS AND PHENOTYPIC CORRELATIONS

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Traditional life history theory is based on a historical natural selection that is defined from the tradeoffs and constraints that have evolved - most likely - from past selection. Examples include i) a fecundity/survival trade-off that selects for Lack's clutch size, ii) a positive correlation between reproduction and body mass that selects for non-negligible body masses, and ii) a ploidy level and mating pattern that define selection on the sex ratio. This approach allows for contingent explanations where some part of the phenotype is explained from other parts of the phenotype. To obtain noncontingent explanations where we predict the joint evolution of life history traits and their trade-offs and correlations we need a deterministic natural selection that is defined exclusively from those constraint and trade-off components that cannot evolve over time. I show how this is possible for Lack's clutch size, large body masses, and non-trivial sex ratios if we focus on the selection pressure that unfolds from population growth and the associated intra-specific and density dependent competitive interactions.

LIFE HISTORY RESPONSE DOES NOT REVEAL FITNESS COST IN LEAD SELECTED DROSOPHILA SUBOBSCURA POPULATION

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Human activities cause severe heavy metal pollution of ecosystems, with lead being one of the most widespread. Heavy metals can have strong effects on survival and developmental time and therefore exert strong selection pressures on exposed populations resulting in increased metal resistance. In populations of different organisms, resistance to heavy metals is generally associated with fitness cost. Evolution of resistance to heavy metals and fitness cost are most efficiently revealed by using laboratory selection protocols. We used laboratory population of Drosophila subobscura selected on elevated concentration of lead for over 30 generations to test if the resistance to lead would show fitness cost. Two laboratory groups, derived from the same Drosophila subobscura natural population, were established. One group of flies was reared on a standard medium (control group) and the other on medium with lead concentration of 100 µg/ml (high concentration of lead - HLC group). Apart from these groups (control and HLC), reciprocal crosses between control lines and HLC experimental group were made in order to investigate maternal effect. The life history traits (viability of all major preadult stages, adult viability and developmental time) of both control and HLC groups and their hybrids were assayed on a standard and a medium with lead. Generally, lead treatment caused lower viability of adults and prolonged the egg-to-adult developmental time. Viability of larvae and adults was lower in hybrids, which also had longer developmental time compared to both experimental groups regardless of the treatment. Our results show that lead concentration of 100 µg/ml did not cause elevated resistance or fitness cost in HLC group. However, the observed significant fitness differences between the hybrids, as well as between the hybrids and the control and HLC groups, suggest the effect of maternal inheritance in resistance mechanism.

WHAT MAKES A GOOD MOTHER? HOW TRADE-OFFS MAINTAIN DIFFERENT MATERNAL INVESTMENT STRATEGIES WITHIN POPULATIONS

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The prenatal environment has important and long-lasting consequences on offspring morphology, physiology, behavior, and ultimately fitness. In birds, many of these prenatal maternal effects are mediated though differential allocation of resources into the eggs. Interestingly, there is large variation in the level of investment among mothers, and little is know why some mother invest little while others invest a lot, and how different maternal investment strategies are maintained within populations. Egg size is highly heritable trait, with known fitness benefits for the next generation. However, in both wild and captive populations, huge variation in egg size is found, with few studies finding evidence for directional selection for larger eggs. By creating replicated selection lines for high and low maternal egg investment in Japanese quail, we show how indirect responses in morphology, physiology and behavior, and sexually antagonistic consequences of selection for high maternal investment constrain the evolution of egg size. Thereby, our study contributes to a better understanding of the mechanisms that maintain variation in maternal investment strategies within populations.

LIFE-HISTORY TRAITS IMPORTANT IN OVERWINTERING: PHENOTYPIC CORRELATION AND GENETIC BACKGROUND

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Adaptation to both annual and long-term changes in environmental conditions is of crucial importance for species living at high latitudes. This involves not only tolerance to cold temperatures, but also an ability to timely predict the forthcoming cold season. Short growing period of temperate zones presents the strongest selection pressure and lead to trade-offs in different life-history traits which are proposed to be regulated by two forecasting mechanisms, circadian rhythms and photoperiodic clock. In several northern species these systems have been found to rely on changes in photoperiod which is the most reliable seasonal cue at high latitudes. We studied four photoperiodically regulated life-history traits, reproductive diapause, cold tolerance, egg-to-eclosion developmental time and juvenile body weight, in a northern fly *Drosophila montana*. Body weight and developmental time were found to be in trade-off regulated by length of photoperiod, and the flies in diapause were more cold tolerant. Induction of photoperiodic diapause is known to be regulated by a photoperiodic clock, while seasonal changes in other traits are governed by circadian rhythms. Life-history traits regulated by these two time measurement systems are likely to respond in different ways to long-term changes in selection regimes, which may break down correlations and create new trade-offs between these traits. To investigate genetic correlation in these traits we performed quantitative trait loci (QTL) analysis. This study revealed both unique QTL (e.g. a large-effect QTL for diapause on the X chromosome), and several combined QTL for these traits (e.g. significant QTL for developmental time and juvenile body, linked with rhodopsin6 gene; combined QTL on the fourth and fifth chromosomes for at least three of the studied traits). The co-occurrence of these QTL suggests that the studied traits are governed partly by the same genes with pleiotropic effects or by multiple tightly linked loci

DISCOVERING LIFE-HISTORY TRADE-OFFS WITH SUPPRESSION OF TOMATO DEFENCE IN THE INVASIVE SPIDER MITE TETRANYCHUS EVANSI

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The herbivorous spider mite Tetranychus evansi is an invasive species specialized to feed on Solanaceous plants, including tomato. In its native range (South America) it is not considered as a pest, but in Africa and Southern Europe major crop losses have been recorded due to T. evansi outbreaks. Apart from predator release, two important factors appear to facilitate the spread of this species: (1) its ability to suppress tomato plant defense to below control levels, and (2) its very high population growth rate. Related species, such as the generalist two-spotted spider mite T. urticae, also have a high population growth rate, but cannot suppress tomato defence to the same extent as T. evansi. Plants protect themselves in various ways against herbivores by producing toxins or attracting natural enemies. Therefore, suppressing these defences can benefit herbivores, and it has been shown that some herbivores indeed do so. However, T. evansi downregulates the defence of its host plant to levels that fall even below house-keeping levels of healthy unattacked plants. Downregulating host plant defence to below house-keeping levels is beneficial for herbivores, but – perhaps surprisingly - not observed before in nature. Given that herbivores do suppress plant defence but only to a limited extent, we hypothesize that suppression of host plant defense trades off with life history traits such as oviposition rate or making dense web to defend a suppressed-defence part of the host plant against competitors. We are currently establishing T. evansi lines that differ in their ability to suppress tomato defense through crossing mites from different geographical locations and subsequent inbreeding, and through experimental evolution. Measurements of life-history characteristics can provide insight into genetic trade-offs for tomato defense suppression. In addition, competition experiments allow us to assess the relative fitness of different T. evansi strategies.

CRAB OSMOREGULATION AND HABITAT DIVERSIFICATION: DOES SALINITY DRIVE PHYSIOLOGICAL EVOLUTION?

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The colonization of fresh water and dry land reflects physiological transformations that govern water and ion movements between the body fluids and surrounding medium, axiomatically linking osmoregulatory physiology and habitat diversification. Since monophyletic groups store biological information retrievable by adding a phylogenetic component to comparative studies, we examine the evolutionary history of osmoregulation in fiddler crabs (Brachyura, Uca). For each osmoregulatory trait examined, we evaluate: (i) phylogenetic pattern, expressed by the relationship between trait variation and phylogeny, employing an autocorrelation analysis using Moran's I coefficient; (ii) the best evolutionary fitting-model, testing Brownian motion (pure random evolution) and Ornstein-Uhlenbeck (random evolution under stabilizing selection) processes; (iii) the ancestral states, to suggest the transformational series, by squared-change parsimony analyses; and (iv) we test the hypothesis of osmoregulatory evolution associated with salinity using phylogenetic regressions. The traits disclosed suggest a directional or piecewise pattern of change over time, revealing strong phylogenetic structuring: for the first pattern, the significant and positive phylogenetic signal among closely related species, particularly at the species level, falls off with phylogenetic distance; for the second pattern, there is a linear change between significant positive and negative phylogenetic signal with phylogenetic distance, showing strong physiological plasticity. The evolution of most osmoregulatory traits follows the Ornstein-Uhlenbeck model: there is significant, strong stabilizing selection that constrains the osmoregulatory history, forcing each trait toward a central value of variation. At more inclusive levels, osmoregulatory ability and salinity are associated, demonstrating a role for both salinity and shared inheritance in driving the evolution of osmoregulatory physiology in fiddler crabs.

INTERACTION OF PARASITE- AND PREDATOR-INDUCED MATERNAL EFFECTS IN THE GREAT TIT (PARUS MAJOR)

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Organisms often face a risk of predation and simultaneously the presence of parasites, which may induce costly defences and thereby strongly influence animal behaviour and physiology. Moreover, if these two types of defences require resources from a common pool, then important trade-offs may arise. Both predation risk and parasites can independently trigger maternal effects to prepare offspring to better cope with their presence in the environment. However, it remains unknown whether the simultaneous presence of both parasites and risk of predation in the maternal environment would induce maternal effects that may affect nestling phenotype, growth rate and condition, and whether the effects induced by the two types of defences are interdependent. We thereby investigated the effects of exposure, pre-laying, during laying and post-hatching, to both presence of parasites and increased predation risk in the great tit and the potential trade-off between them. In increased predation risk environments, the maternal response enhanced wing growth and immune response of male nestlings. Nestling mass, tarsus and survival were influenced by parasites, but their negative effects were attenuated by the maternal response. The effects of the predator treatment on mass seemed to be concealed by a stronger effect of parasites, but were visible on tarsus growth. The findings suggest that maternal effects induced by an increased perceived risk of predation and by parasites influence offspring phenotype and survival, that the two stressors interact with each other, and that their simultaneous presence can alter the optimal allocation of resources mediated by maternal effects. The presence of parasites seems to have a stronger effect on growth and development compared to that of predators, suggesting a prioritization of the different risks in a particular environment.

EXPERIMENTAL MANIPULATION OF REPRODUCTIVE COSTS DURING THE PRE-BREEDING PERIOD REVEALS CONDITION DEPENDING TRADE-OFFS IN A LONG-LIVED, SOCIAL BIRD SPECIES

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Life-history theory predicts that lifespan is a key predictor of parental investment decisions. A key tool used to investigate parental investment trade-off in birds are clutch-size manipulations, revealing that an increased parental effort reduces survival of male but not female breeders. However, it remains often unclear if intra-generational costs are traded in against inter-generational costs (i.e. if parents pass on costs of increased reproductive investment to their offspring or pay the cost themselves). Here we investigate this core trade-off of life-history theory by experimentally lowering the condition of female breeders before the onset of the breeding season in Siberian jays (Perisoreus infaustus), a long-lived bird species where some of the offspring remain with their parents for up to three years. We injected female breeder with brucella abortus (BA) solution, which lowers the condition of birds for 2-3 weeks, or saline solution in two years with contrasting climatic conditions during the breeding season. BA injections had no effect on any breeding parameter in a favourable year with high breeding success. In an unfavourable year, however, BA injected females laid fewer and smaller eggs, and consequently, only fledged in average 0.30±0.30 offspring, compared to 2.5±0.36 offspring in saline injected females. We found no difference in offspring quality, and subsequent survival of retained offspring and females across the treatments and years. Thus, females in long-lived bird species can reduce the number of offspring under unfavourable conditions, but still produce high quality offspring. These results demonstrate the value of evolving variability in life-history traits in unpredictable environments.

DOES INCREASED STRESS RESISTANCE REDUCE THE ABILITY TO DEAL WITH BIOTIC CHALLENGES? A TEST THROUGH SELECTION EXPERIMENTS ON DROSOPHILA MELANOGASTER

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Humans impact ecosystems in a multitude of ways, increasingly exposing contemporary organisms to abiotic and biotic stressors. Environmental stress has strong negative impacts on biological diversity, as species can go locally extinct, if they are unable to migrate to a more benign habitat or to overcome the stressor via plastic or evolutionary adaptation. Several factors are thought to constrain adaptive evolution, such as gene flow, lack of genetic variation and genetic or functional trade-offs. Despite trade-offs being postulated as playing a central role in evolutionary theory, interactions between abiotic and biotic stress resistances have rarely been investigated in stress adaptation. This project aims to test for trade-offs between abiotic and biotic stress resistances in the well-established Drosophila melanogaster study system. Selection experiments on stress resistance provides an opportunity to study evolutionary constraints resulting from genetic trade-offs between traits. Lines that have been selected for different abiotic stressors (heat, cold and desiccation) will be tested for costs and benefits, whereas performance will be tested in outdoor cages under different biotic stressors (competition, predation, parasites) and under different climatic conditions (hot, moderate and cold days). These experiments provide a test of whether trade-offs between abiotic and biotic stress resistances are potential constraints to stress adaptation, which is crucial to better understand the evolutionary potential of contemporary populations.

EXTERNAL VS. INTERNAL IMMUNE DEFENCE IN THE RED FLOUR BEETLE (TRIBOLIUM CASTANEUM)?

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The red flour beetle, Tribolium castaneum, secretes quinones that control the microbial flora in the surrounding environment. These secretions act as an external immune defense that provides protection against pathogens. At high concentrations, however, these secretions are harmful to the host itself, and selection may thus have optimized the level of expression under natural conditions. Here we show that the expression of external immunity responded to selection during experimental evolution within a few generations. At the same time, one component of internal immune defense (phenoloxidase activity) was compromised in beetles selected for either high or low external defenses. Protection against a natural pathogen was lacking in flour obtained from beetle lines selected for low amounts of secretions. Altogether, this suggests that external and internal immune defenses work together efficiently under natural conditions, while every manipulation on the side of external immune defense comes with costs to the internal immune defense.

EVOLUTION OF MALE REPRODUCTIVE AGEING UNDER DIFFERENTIAL RISK OF DEATH: THE ROLE OF CONDITION-DEPENDENT MORTALITY

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Ageing is observed in most organisms and is an important aspect of their life histories. However, how ageing evolves is not fully understood. The classic evolutionary theory of ageing posits that high extrinsic mortality leads to evolution of rapid ageing and therefore shorter intrinsic lifespan. On the other hand, according to a novel theory, when mortality is condition-dependent and surviving cohort is not a random sample of the starting population, which is likely to be the case in nature, the classic prediction does not hold and increased mortality can result in postponed onset of ageing. To increase our understanding of how different types of extrinsic mortality may influence the evolutionary trajectories of ageing, we have previously conducted an experimental evolution study, using the nematode *Caenorhabditis remanei* as a model system. This study allowed us to disentangle the effects of mortality rate (High and Low) and mortality source (Condition-dependent and Random) on the evolution of intrinsic lifespan. We observed reduced longevity under high random mortality, in accordance with the classic prediction. However, when mortality was condition-dependent, high mortality resulted in the evolution of increased lifespan supporting the novel theory and highlighting the importance of mortality source. Surprisingly though we found that there was no trade-off between longer lifespan and reproduction in females. In the present study we examined the possible existence of such trade-off in males. We found that males evolving under condition-dependent mortality had lower reproductive performance, suggesting a trade-off between stress resistance and reproduction. However, these males enjoyed a lower rate of reproductive ageing than males evolving under random mortality. Our study suggests that condition-environment interactions play a key role in evolution of male reproductive ageing.

FITNESS EFFECTS OF A TRANSPOSABLE ELEMENT INSERTION DEPEND ON ENVIRONMENTAL CONDITIONS IN DROSOPHILA MELANOGASTER

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Understanding how adaptation works is one of the major questions in Evolutionary Biology. Towards answering this question, we identified recent Transposable Element (TE)-induced adaptations in Drosophila melanogaster and we are currently connecting them to their molecular mechanisms and fitness effects. Specifically, the TE insertion named Bari-Jheh, is present at high frequency, but not fixed, in all the populations analyzed and shows signatures of a selective sweep. *Bari-Jheh* is inserted in the intergenic region between Jheh2 and Jheh3 genes encoding for Juvenile Hormone Epoxy *Hydrolase*. Previous results show that *Bari-Jheh* is associated with a down-regulation of the expression of its nearby *Jheh2* and *Jheh3* genes. Furthermore, *Bari-Jheh* is also associated with reduced viability and extended developmental time that probably represent the cost of selection of this insertion. In order to identify the adaptive effect of this insertion, we analyzed the sequence of *Bari-Jheh* and we found that it contains two Antioxidant Response Elements. This suggests that the presence of the insertion could lead to an increased level of expression of Jheh genes under Oxidative Stress (OS) conditions and to increase resistance to this stress. We indeed found that under OS conditions the TE is associated with an increased expression of its nearby genes *Jheh1* and *Jheh2* (t-test p< 0.05) and that flies with the insertion show a higher survival rate compared to flies without the insertion (Kaplan-Meier logrank test: p<< 0.001). Our results show that the fitness effects of TE insertions could depend on the environmental conditions. This would explain why adaptive TEs such as *Bari-Jheh* are present at high frequencies but not fixed in any population. Moreover, this study shows that we need to explore several phenotypes in order to fully characterize the effects of an adaptive mutation.

RECONCILING OVERALL VIRULENCE WITH VIRULENCE EVOLUTION UNDER CONDITIONS OF FREQUENT MULTIPLE INFECTIONS: INSIGHTS FROM THE DAPHNIA-PASTEURIA HOST-PARASITE SYSTEM

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Free-living organisms are regularly found to be infected by an assemblage of genetically distinct parasite strains. Despite the ubiquity of multiple infections in a variety of organisms including humans, our current understanding of the processes driving virulence evolution is limited. This is because empirical studies using animals involve single generation experiments which measure the expression of virulence in multiply-infected hosts (overall virulence), whereas theoretical approaches model the evolution of virulence by studying how within-host competition affects between-host transmission (virulence evolution). More importantly, few empirical studies assessed the relative strengths of ecological and epidemiological feedbacks, and the associated trade-offs among fitness components of coinfecting parasite strains. Here I present recent results from two studies of multiple infections and the evolution of virulence using the Daphnia-Pasteuria host-parasite system. This system is ideal for studying virulence evolution at the organismal level, because host clonal reproduction can be controlled in the laboratory, and the relative contribution of parasite clones (single genotype) during multiple infections can be reliably quantified. I will show that parasite within-host competitiveness does not always correlate with host castration ability. I will also show that overall virulence does not correlate positively with parasite infectivity and transmission. More importantly, the exact relationship among these parameters crucially depends on host age. Finally, I will show that under conditions of frequent multiple infections, the virulence-transmission trade-off may become one of several trade-offs faced by coinfecting parasite strains. Taken together, these results stress the need to conduct long-term studies of virulence evolution using multiply-infected hosts.

METABOLIC RATE AS A PROXIMATE MECHANISM UNDERLYING LIFE HISTORY STRATEGIES IN FLYCATCHERS

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Competitive exclusion can occur when closely related species inhabit the same geographic area, unless there is some type of niche differentiation. Collared and pied flycatchers are closely related sympatric passerine species that compete for nesting sites and other resources. These species appear to have solved the life history trade off between competitive interference and robustness to poor environments differently. Collared flycatchers experience an advantage in competition over breeding sites while pied flycatchers are more robust to poor conditions. These different life history strategies may promote regional coexistence, but proximate mechanisms underlying these strategies are unknown. We will test if the differences between pied and collared life history strategies are associated with differences in metabolic rate. Higher metabolic rate has previously been associated with higher competitive ability, faster growth rate and more resource use, traits that differ between collared flycatchers and pied flycatchers. Resting metabolic rate (RMR) of collared, pied and hybrid flycatchers, both chicks and adults, will be measured using respirometry and combined with brood size manipulation experiments during the breeding season on the Swedish island of Öland in 2013. We will test if metabolic rate is significantly different between species, specifically whether collared flycatchers have higher metabolic rates than pied flycatchers. And how metabolic rate is related to growth under different environmental conditions (experimentally manipulated through brood size changes). We will also determine if metabolic rate is associated with growth rate and territory quality within and between species. Understanding the metabolic rate of closely related, hybridizing species could give insight into proximate mechanisms that facilitate life history strategies and regional coexistence.

BIG HOUSES, BIG CARS, SUPERGUPPIES AND THE COSTS OF PRODUCING SPERM

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Trade-offs between fitness related traits are expected to generate negative genetic covariances, unless genetic variance for resource acquisition exceeds that for resource allocation. For what concerns postcopulatory sexual selection, producing competitive ejaculates is costly, however, and it can be attained at the expenses of traits involved in mate acquisition and/or survival, because resources are limited. Alternatively, under a good-sperm scenario, males that are better equipped for winning sperm competition should also show higher survival and, more generally, perform better than poor sperm competitors. In the guppy (*Poecilia reticulata*) the number of sperm inseminated is the most important determinant of male fertilization success, therefore to evaluate the evolutionary consequences of male investment in sperm production we performed a bidirectional artificial selection experiment for sperm production in guppies. Contrary to expectation, males selected for high sperm production not only transferred more sperm per copulation, but also grew faster, attained a larger body size, were more colourful, sexually more active and more attractive to females. Sperm velocity and morphology was not affected, but sperm viability increased with sperm number. All the other measured male and female fitness-related traits were unaffected by sperm production. These results suggest that, at least in lab conditions, some guppy genotypes perform better in most life-history traits, acquiring more resources than others.

LIFE HISTORY AND REPRODUCTIVE TRADE OFFS IN CAECILIAN AMPHIBIANS

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The study of trade-off mechanisms is particular interesting in animal group showing extraordinary variations of life history strategies. Among vertebrates, snakelike caecilian amphibians (Gymnophiona) exhibit a considerable variation of reproductive modes including both oviparity and viviparity combined with highly unusual investment strategies (e.g. skin-feeding and intrauterine feeding). We integrated the current knowledge on reproductive traits and life histories into a comparative multidimensional analysis to understand the scenario behind the evolution of reproduction in this group. Our results are compared with the available life history data of other amphibian lineages representing other body forms and phenotypes.

LONGEVITY AND REPRODUCTIVE PERFORMANCE STRATEGIES

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Reproductive performance differs between individuals depending on their condition status and individual quality. It is also shaped by external factors and, moreover may change with progressing age. Both within-individual changes and lifetime reproductive strategies differ between species and one factor shaping it is longevity. Short- and long-lived species are subjected to different constraints influencing not only their reproductive strategies but even possibilities for these strategies. I have analyzed possible mechanisms behind age-related changes in reproductive performance and lifetime reproductive success (LRS) in two passerine species. The short-lived Collared flycatcher (*Ficedula albicollis*) shows strong selection against poor breeders at early ages and quality/longevity-dependent LRS. LRS in the long-lived Siberian jay (*Perisoreus infaustus*), is dependent on social status and at least some individuals seem to be able to compensate for initial lower quality by delaying reproduction. Thus, the available strategies of shaping reproductive performance appear to differ between species dependent on their longevity.

PHENOTYPIC AND GENETIC CORRELATIONS FOR SEVERAL POTENTIALLY INTERACTING TRAITS IN MEDITERRANEAN MUSSEL (MYTILLUS GALLOPROVINCIALIS)

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The genetic architecture of several traits whose response to selection could be correlated was carried out by studying their genetic and phenotypic correlations in several cultured families of Mediterranean mussel (Mytilus galloprovinciallis).Growth traits (weight and length at two years of age), hepatopancreas accumulation of Diarrhoeic Shellfish Poisoning (DSP) toxins from plankton) and mantle colour were studied. All these traits are important from the point of view of the commercial production of this species. Although the toxins evaluated in this study are known not to kill individuals, it is possible that, to some extent, their development could be affected by toxin accumulation. To inspect this, we evaluated mussel growth and hepatopancreas toxin concentration in 2500 offspring from a set of 280 potential families obtained by crossing 10 females x 28 males in controlled conditions. After hatching and metamorphosis in lab conditions, mussels were wrapped on ropes hanging from production rafts and exposed to a natural bloom of toxic algae. Phenotypes were measured at individual level including length, weight, colour, and toxin amount and concentration. Using a microsatellite parentage tool, close to 90% individuals were assigned to a specific couple. Some individuals from wild settlement were also detected. With this information phenotype and genotype correlations were obtained for the evaluated traits. For the group of individuals we have analyzed, response to directional selection would be expected for growth, since we have estimated its narrow-sense heritability to be significantly different from zero. Regarding toxin accumulation, we obtained low estimates of narrow-sense heritability. We discuss the implications of the results in terms of a putative trade-off between both traits.

NEITHER FEMALE REPRODUCTIVE COSTS NOR MALE GONADAL ANDROGENS DRIVE DEVELOPMENT OF SEXUAL SIZE DIMORPHISM IN LIZARDS

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What developmental mechanism allows substantial evolutionary plasticity in sexual size dimorphism (SSD) among squamate reptiles (lizards and snakes)? Currently, the most pervasive candidates are the opposite effects of testicular testosterone on growth in male-larger versus female-larger species and the differences between sexes in energy allocation to growth versus reproduction. We tested these alternatives by manipulating testosterone levels and reproductive status in the male-larger lizard Paroedura picta. Males reached comparable final body length regardless of circulating testosterone levels. We conclude that previously reported effects of gonadal androgens on growth in male lizards may have been a consequence of altered behaviour in manipulated individuals. Non-reproducing females in social isolation reached the same final body size as did females that frequently reproduced, indicating that division of energy into growth versus reproduction is not the mechanism generating SSD. Notably, ovariectomized females and females with exogenous testosterone grew to larger body size than did intact females. These findings demonstrate that ovariectomy causes adverse side effects and is not a suitable technique for removing female reproductive costs. We argue that our results and the published evidence are in accordance with endogenous control of SSD in squamates by female gonads independently of direct energy allocation to reproduction.

A SINGLE MOLECULE ASSAY TO STUDY THE FITNESS EFFECTS OF LEAKY GENE EXPRESSION IN INDIVIDUAL CELLS

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Most of the adaptive substitutions that are revealed in evolution experiments are in regulatory sequences. In many cases, the expression levels are under directional selection. The *lac* operon in Escherichia coli is a good model system to investigate the fitness effects of the different regulation precisions (tight or leaky regulation) under different conditions. LacI, the repressor of the *lac* operon, can bind to three different operators with different binding strengths (O1>O2>O3). Yet, LacI can bind even stronger to the artificial operator sequence, Osym. Thus, the evolutionary optimal case may not necessarily be the tightest regulation of the *lac* operon. Our hypothesis is that the individuals with *lac* operon leakage will have a fitness disadvantage due to the cost of the unnecessary expression of *lac* operon when there is no lactose to utilize, but if lactose suddenly becomes available those individuals will benefit from previous leakage and have a higher fitness than the individuals with no leakage. Bulk growth assays can only assess fitness differences that are due to differences in genotype. Unless one looks at the single cell level it is not possible to reveal the contribution of stochastic aspects of the phenotype to observed fitness differences between isogenic populations. We do this by correlating leakage events in the lac operon of an individual *E. coli* in lactose free medium, with its fitness before and after the switch to lactose medium. The leakage events are quantified using single molecule fluorescence microscopy where we can count the number of lactose permease molecules per cell (Choi 2008). The leakage in expression is monitored in cells growing in a microfluidic turbidostat (Ullman 2013). Here, we can follow growth of thousands of individual cells with high time resolution and switch between different growth media. This enables us to relate minute differences in fitness to the underlying stochastic differences in gene expression.

Symposium

21. The Evolution and Maintenance of Heritable Colour Polymorphsims: from Ecology to Genomes

20 and 21 August



Program

Tuesday 20 August Session(s): 4 Wednesday 21 August Session(s): 5, 6

Organisers: Maren Wellenreuther and Bengt Hansson

Invited speakers: Rosemary G. Gillespie, Jon Slate Chris D. Jiggins

Description:

Heritable colour polymorphisms are widespread in nature but the processes governing the maintenance and evolution of different morphs in the wild are rarely well understood. The field of polymorphism research is currently progressing rapidly because of the fruitful integration of ecology and genetics with bioinformatics and genome technology. This advancement allows researchers to now address questions about evolutionary processes of polymorphisms, such as the genomic patterns of selection and the functional consequences of genetic architecture.

D21SY21IT10:30R2

THE ROLE OF TRANSIENT VERSUS BALANCED COLOR POLYMORPHISM IN ADAPTIVE RADIATION: SPIDERS IN THE HAWAIIAN ISLANDS

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Color variation, when genetically determined, provides a visual tool for examining selection. Moreover, variation is a key to rapid adaptive response, which in turn forms the basis for adaptive radiation. Our work focuses on different genera of spiders in the Hawaiian Islands, in particular: (1) Long jawed spiders in the genus Tetragnatha (Tetragnathidae) that are characterized by exuberant adaptive radiation. This genus has colonized the islands an estimated 5mya and shows evolutionary progression from older to younger islands. One lineage, the "spiny leg clade", is represented throughout the archipelago by four distinct color ecomorphs and molecular phylogenetic analyses indicate that each ecomorph has evolved repeatedly. However, species on the older islands (Kauai and Oahu) show a developmental switch between two ecomorphs - Green and Maroon. More derived species on the younger islands exhibit a single ecomorph, Green or Maroon, at all life stages, though species formation is associated with multiple shifts between ecomorphs. We are currently using a genomic approach to understand how the color-switching characteristic of species on the older islands has translated into diversification of species representing different ecomorphs on the younger islands. This system contrasts with (2) the exuberantly patterned Hawaiian Happy Face spider, Theridion grallator, a single species which displays a visible and balanced genetic color polymorphism. The happy face spider retains variability between individuals within a population (as compared to within individuals or between species in Tetragnatha). Moreover, while the frequency of color morphs is similar in different populations of happy face spiders, the mode of inheritance of the color polymorphism has changed between islands in the Hawaiian chain. Together, these studies provide insights into how, and under what circumstances, variability can translate into diversity.

D21SY21IT14:00R2

THE ORIGINS OF TWO COLOUR POLYMORPHISMS IN A WILD MAMMAL POPULATION

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In a wild population of Soay sheep, that has been the focus of a >25 year field study, two coat colour polymorphisms are segregating as single locus Mendelian traits. Previously, the underlying mutations have been discovered, and it has been shown that both genes are under selection. However, the age and origins of the polymorphisms were unknown. In this talk, we show that Soay Sheep underwent admixture with more modern breeds in the 1800s, and that both polymorphisms arose through the introgression of domesticated genetic variants into the Soay population. Therefore, the current field site can be regarded as a 'natural laboratory' in which wild type and domesticated variants have been competing with one another for over 100 years. It is shown that domesticated and wild populations may be a common source of genetic variation, capable of providing the material required for evolutionary change and adaptation.

D20SY21RT17:45R8

ECOLOGY AND MATING INTERACTIONS: TEMPERATURE INFLUENCES MALE-FEMALE CONFLICT IN A COLOUR POLYMORPHIC DAMSELFLY

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Heritable and conspicuous colour polymorphisms have a long research tradition in ecological genetics, and these systems have been used to investigate issues such as negative-frequency-dependent selection (NFDS), maintenance of genetic variation, sexual selection and sexual conflict. Here I will present long-term field observational data and experiments on the evolutionary dynamics of a sexually selected colour polymorphism in the damselfly Ischnura elegans. Three female morphs exist in this species, one of them being a male mimic ("androchrome females"). Androchrome have lower mating rates than other female morphs, suggesting that male mimicry is a female defence against excessive and costly male mating harassment that is detrimental to female fitness. I will present long-term field data from a longitudinal study across multiple populations that show the stability of this female polymorphism and the results of experiments where we have manipulated morph frequencies and densities and evaluated the effects on morph and population fitnesses. I will also present data showing that the male-female mating interactions are environment-dependent and moulded by ambient temperatures, resulting in geographic variation in morph frequencies. Thus, sexual conflict in this system and the benefits of male mimicry is highly context-dependent upon local ecology and the thermal environment.

D20SY21RT18:09R8

NATURAL SELECTION DRIVES INCIPIENT DIVERGENCE DESPITE CONTINUOUS GENE FLOW IN SYMPATRIC COLOR MORPHS OF A CORAL REEF FISH

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The role that natural selection plays in initiating divergence-with-gene-flow remains controversial. Color polymorphisms offer rich opportunities to explore how selection can drive the evolution of reproductive isolation even in the face of gene flow. We integrate genetics, ecology, and behavior to outline how disruptive selection on color pattern in combination with assortative mating is driving divergence in sympatric color morphs of the arceye hawkfish (Paracirrhites arcatus) in Hawaii. First, ecological surveys show a strong correlation between phenotype & environment. Fine-scale microhabitat variation in visual backgrounds creates distinct niches that appear to favor alternative color patterns. Experimental tests are ongoing, but preliminary evidence shows reduced fitness in morphs found in contrasting habitats. Second, field observations of mating pairs indicate that, even in zones of overlap, fish are 10x more likely to pair with like morphs. Lab-based experiments of captive fish are currently being used to further explore the strength of assortative mating and its impact on reproductive isolation. Third, genome scans using 30 microsatellite loci show divergent patterns of genetic variation across morphs indicative of divergence-with-gene-flow; and outlier detection methods confirm three loci as candidates for positive selection. The strong association between genetic variants, color pattern, and ecological gradients all suggest that P. arcatus has evolved genetically-based alternative phenotypes that are adapting to spatially heterogeneous habitats despite continuous gene flow. We are currently using comparative genomics to discover regions showing signatures of natural selection and identify the underlying genetic basis of adaptive color pattern and correlated traits. This research outlines one of the few case studies of ongoing sympatric divergence in any marine fish and will help elucidate the role color polymorphism plays in promoting speciation.

D20SY21RT18:33R8

EXPERIMENTAL AND GENOMIC APPROACHES IN THE STUDY OF THE BALANCED COLOUR-POLYMORPHISM OF THE MEADOW SPITTLEBUG (PHILAENUS SPUMARIUS)

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Philaenus spumarius (Insecta, Hemiptera, Aphrophoridae) has for long been a subject of interest of evolutionary biologists due to its heritable colour polymorphism that shows evidence of balancing selection and of clinal variation in the colour mophs frequencies. We are studying the adaptive significance of this polymorphism, particularly to understand if the melanic morphs (e.g. "marginellus" morph) have any advantage/disadvantage in terms of survival and reproductive success, efficiency of egg maturation and resistance to desiccation compared to non-melanic morphs ("typicus" and "trilineatus"). Results so far indicate a higher survival, higher number of eggs clutches and higher number of eggs laid by the "trilineatus" females than "typicus" or "marginellus" females. We are also taking a genomic approach for a) the identification of genetic basis of the colour polymorphism and b) for detecting signatures of balancing and directional selection in the genome of *P. spumarius*. For this purpose we are applying RAD sequencing in a) a set of samples from the three different morphs referred above, using a high frequency cutter enzyme (PstI) and in b) another set of samples from 8 populations across the distribution range of the species representing the main mitochondrial haplogroups, using a lower frequency cutter enzyme (SbfI). We are also assembling a draft of the genome that will aid in the identification of homologous regions to available references, although the very large genome size of this insect constitutes an extra challenge.

D20SY21RT18:57R8

GENETIC AND GENOMIC INSIGHTS INTO A COLOUR POLYMORPHISM OF THE GOULDIAN FINCH (ERYTHRURA GOULDIAE)

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The *Red* locus of the Gouldian finch *Erythrura gouldiae* is a pigmentation switch that determines black (melanin) and red (carotenoid) morphs. This locus offers a unique opportunity to investigate the genetic basis and evolutionary history of a sympatric polymorphism for colour that has also been found to be associated with multiple physiological and behavioural differences. We used classical linkage mapping combined with RAD (Restriction site Associated DNA) sequencing and association analysis to localize the locus to a small (65-kb) genomic region. The pattern of nucleotide diversity at the *Red* locus is characterized as a genomic island that shows significant differentiation and divergence between the black and red haplotypes. Evolutionary theory suggests two distinct hypotheses to explain this pattern. First, sequence divergence may have accumulated within the species due to reduced gene flow between morphs, supported by the observed high degree of pre- and post-zygotic incompatibility, or due to a genomic rearrangement. Second, the sequences may have diverged in isolated lineages, prior to population merger or introgression. We examined the two alternative possibilities by using genetic and genomic tools and assess the evidence for balancing selection.

D21SY21RT11:18R2

KING MIDAS MEETS MENDEL: UNRAVELING THE GENETIC BASIS OF AN EVOLUTIONARILY RELEVANT COLOR POLYMORPHISM

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Color polymorphisms are tractable traits that are directly related to various crucial evolutionary processes including adaptation and speciation. The genetic basis of coloration has been the focus for genetic and evolutionary research for over a century. Despite this interest, few studies have successfully unraveled the genetic basis of color polymorphisms in natural populations; and the processes governing the origin, spread and maintenance of polychromatism are still poorly understood. The Midas cichlids of the crater lakes in Nicaragua are an excellent system to investigate the maintenance of a conspicuous color polymorphism and its role in speciation-with-gene flow. Midas cichlids are characterized by a conspicuous polychromatism: most individuals are grayish with dark bars, while some exhibit a gold coloration. The color morphs mate assortatively and have been suggested to be undergoing divergence in sympatry. Here, we explore the genetics of this trait using a combination of genetic mapping and next-generation sequencing technologies. The results show that a simple mendelian genetic architecture can underlie sympatric divergence, and might even constitute evidence for one-gene models of speciation. We further show that the onset of the phenotype is variable and dosage dependent (i.e homozygotes have earlier onset). This observation is of interest to understanding the fitness landscapes under predation pressure in the wild. The genomic region that harbors the gold locus is genomically unstable and characterized by the presence of tandem duplications and innumerous indels. The genes in the region are related to immune function, coloration and social affiliation. Patterns of association found in population-based fine-mapping using SNP genotyping and targeted-enrichment NGS are complex and compatible with multiple origins. These results indicate a role of mutation rate, hitch-hiking and pleiotropy in the maintenance of this conspicuous color polymorphism.

D21SY21RT11:42R2

SELECTION ON FLOWER COLOUR GENES IN A SNAPDRAGRON HYBRID ZONE

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A major goal of evolutionary biology is to understand how diverging populations become distinct species. Although much progress has been made in identifying genes that contribute towards population divergence and speciation, direct measurements of their effect on fitness in nature are often difficult to obtain. We are studying the evolutionary dynamics of speciation between two subspecies of Antirrhinum (snapdragons) with different flower colours (yellow and magenta). In this system, two major loci control flower colour and give rise to six colour phenotypes across a narrow hybrid zone. Surprisingly, these species are visited by the same array of pollinators (large bees). This raises the question of how these genes initially arose and spread and how populations are currently maintained in the face of gene flow. To better understand the role of flower colour genes in speciation we are using an integrated approach to examine the role of selection, epistasis, drift and gene flow over multiple time scales. To examine short and long-term evolutionary processes we are using SSR and SNP markers for individuals sampled across the hybrid zone as well as sequence variation with Restriction-Associated DNA sequencing (RAD) data from allopatric populations. The existence of a steep cline (~280m wide) for flower colour and diagnostic markers linked to the underlying genes, suggests selection is acting against some of the hybrid colour phenotypes. Significant heterozygote deficit at small spatial scales (<30m) and an excess of parental phenotypes also suggests assortative mating may play a role in maintaining the hybrid zone. Initial results from RAD tags indicate that the highest regions of divergence (Fst) between allopatric populations are located near the genes that control flower colour. Taken together, these results suggest that selection is acting on the flower colour genes, likely through pollinators discriminating among the flower colour phenotypes within the hybrid zone.

D21SY21RT14:48R2

DEFINING THE REGULATORY REGIONS THAT CONTROL HELICONIUS BUTTERFLY COLOUR PATTERN MIMICRY

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Genomic studies of natural populations are offering novel insights into adaptation and diversification. In particular, recent studies of parallel evolution of similar phenotypes in divergent lineages have commonly shown the utilization of shared genetic variation. *Heliconius* butterflies represent a recently documented example of shared allelic variation across species boundaries. Heliconius display bright wing patterns that warn predators of distastefulness and also act as mating cues. The diversity of patterns displayed within and between the hundreds of forms is remarkable, as is the convergence between species onto near-perfect mimetic patterns. Recent field studies have identified a group of populations along the eastern slopes of the Andes that are allied to *H. timareta* and share wing phenotypes with sympatric *H. melpomene*. Genomic studies have shown that the populations with similar phenotypes also share allelic variation at wing patterning loci, with adaptive introgression across the species boundary providing the most likely explanation for this pattern. We sequenced a 600 Kb genomic region that regulates diverse red wing pattern phenotypes, using 80 Heliconius samples. Genomic intervals associated with at least three independent red colour pattern phenotypes were resolved using sequence comparisons that grouped similar wing phenotypes, irrespective of species. By comparing the level of nucleotide variation within each colour pattern interval, we estimate the time in generations when introgression events occurred between *H. melpomene* and *H. timareta*. Gene exchange after speciation has resulted in the adaptive spread of colour pattern alleles. Here we have identified narrow genomic regions that must act through cis-regulatory control of the transcription factor *optix*, in order to control complex phenotypes.

D21SY21RT15:12R2

SPECIATION GENOMICS IN THE EUROPEAN CROW: A MAGIC HYBRID ZONE WHERE SEXUAL SELECTION AND FEW MAJOR EFFECT LOCI MAY PROMOTE SPECIATION

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Plumage colour differences within and between species are characteristic of many organisms and are particularly conspicuous in birds. The evolution of such colour polymorphisms, their contribution to prezygotic isolation and their general role in speciation has been a central theme in the evolutionary sciences. The hybrid zone between carrion and hooded crows (Corvus [c.] corone and C. [c.] cornix) is a prime example. The zone appears to have been maintained by strong assortative mating mediated by plumage colour for at least a century. The discrete segregation in colouration strongly contrasts with a surprising lack of genetic differentiation. Here, we aim at identifying the genetic basis of this colour polymorphism and assess its role in the speciation process. To achieve this goal, we generated an annotated draft reference sequence of one Hooded crow individual which we then used a backbone for population genomic analyses. We further conducted an extensive (RNAseq) gene expression experiment under common garden conditions focusing on differential gene expression of genes expressed in active feather follicles. The combination of the population genomic data and detailed information from the gene expression experiment provides a powerful approach to identify the genetic cause of colouration differences and its consequence for local genomic divergence between this pair of incipient species.

POSTERS

D21SY21PS0059

MAINTENANCE OF FLOWER COLOR POLYMORPHISM IN IRIS LUTESCENS LAM. (IRIDACEAE)

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As pollinator visitation rate is generally correlated with plant fitness in entomogamous species, stabilizing selection mediated by associative learning is expected to occur on floral traits, leading to low intra-specific variation. Flower color is among the most important visual signals to attract pollinators. Consistently, previous studies suggested that flower color polymorphism could be maintained by pollinator-mediated selection. In addition, flavonoid/anthocyanin, carotenoid pigments, which are responsible for coloration in most plants, have pleiotropic effects for plant survival. Therefore selection pressures induced by a range of abiotic (precipitation, soil or temperature) /biotic (herbivores and pathogens) factors could also be involved. Herein, we investigated flower color polymorphism in a common species in the Mediterranean area, *Iris lutescens* Lam. (Iridaceae), which displays a tremendous purple-yellow flower color polymorphism within populations. We revealed that I. lutescens is a rewardless species displaying mainly different anthocyanin accumulation induced flower color difference, that is not associated with marked difference in odor emitted nor level of defense against abiotic/biotic factors. Results of pollinator/florivore observations and genetic analyses underlying flower color variation will also be presented in this talk. Pollinators are the most probable selective agent responsible for the maintenance of this polymorphism, suggesting that this species is a good candidate to investigate the maintenance of flower color polymorphism.

D21SY21PS0208

FLORAL PIGMENTATION EVOLUTION AND MOVEMENT THROUGH GENETIC SPACE IN SNAPDRAGONS (ANTIRRHINEAE)

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Floral pigmentation is a conspicuous and variable trait with an enormous effect on sexual selection, and the underlying genetic pathways are well-understood. As such, flower colour evolution has great potential to inform our understanding of the relationship between genes and phenotype. Previous work has found consistent patterns of irreversible transitions from purple to white, red and blue flowers, via single-locus loss-of-function mutations in regulatory genes of the flavonoid pathway. Since yellow pigmentation is typically controlled by a wholly different pathway, transitions to yellow must involve mutations at two loci. However, transitions involving yellow flowers, or at broader taxonomic scales have never been properly examined. I present results from an ongoing comparative study of floral evolution in a tribe of snapdragons (Antirrhineae, Plantaginaceae), which aims to assess patterns of evolution between yellow, white and purple flowered species in a phylogenetic context. Single locus transitions appear to dominate at short time scales, demonstrating the need to look beyond the genus level. Transitions also appear to be generally more reversible than in previously examined groups. I also examine the role of other floral traits that may be correlated with shifts in flower colour.

D21SY21PS0274

PREY AND PREDATOR COMMUNITY COMPOSITION PROMOTES POLYMORPHIC WARNING SIGNALS

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Polymorphic warning signals are puzzling since positive frequency-dependent selection should promote monomorphic warning coloration. We studied predation pressure in the aposematic moth *Parasemia plantaginis* by using artificial prey resembling white and yellow male colour morphs in five separate populations. We tested if predation was influenced by: 1) natural frequencies of colour morphs; 2) number of interspecific Lepidopterans sharing similar coloration, and; 3) predator community composition. Predation on yellows was lower than whites' regardless of their local frequency. The number of white interspecifics increased the attack risk of whites and decreased it on yellows, whereas yellow interpecifics lowered predation on both morphs. Interestingly, predation pressure was dependent on predator community composition: Yellows suffered less attacks when Paridae were abundant, whereas whites suffered less attacks when Prunellidae were abundant. Our results suggest spatial heterogeneity in prey and predator community composition can generate geographic mosaic selection facilitating the evolution of polymorphic warning signals.

D21SY21PS0514

THE GENETIC BASIS OF ADAPTIVE PLUMAGE COLOUR POLYMORPHISM IN COMMON BUZZARDS

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Common buzzards (Buteo buteo) display substantial variation in ventral melanin-based plumage colour, with three main morphs. Intermediate morph birds have higher lifetime reproductive success than pale and dark morphs in a German study population. Transmission studies suggest that a major locus underlies much of the colour variation. Here we investigate genotype-phenotype association at the MC1R locus, which has been frequently implicated in melanin polymorphisms in birds and other vertebrates. We find significant association between several SNPs and morph colour, with the strongest association occurring in a region upstream of the MC1R coding region. No association is found at other loci, including the ASIP pigmentation locus. These results imply that regulatory mutations at MC1R underlie variation in adaptive plumage coloration in buzzards, which provides a strong contrast to other studies in vertebrates to date, which have focused on the MC1R coding sequence. Thus a further nuance in the issue of the relative importance of coding and regulatory mutations in adaptation is that both types of mutation can occur at the same locus.

D21SY21PS0567

INVESTIGATING THE ROLE OF DOMINANCE IN THE POLYMORPHISM OF WING COLOUR PATTERNS IN THE MIMIC BUTTERFLY HELICONIUS NUMATA

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Dominance is a widespread mechanism by which the phenotype of heterozygotes is determined. In polymorphic loci, dominance thus plays an important role in the dynamic of the phenotypic polymorphism because of the high number of heterozygotes. This talk focuses on dominance in a striking case of colour polymorphism caused by Müllerian mimicry. In the unpalatable butterfly species Heliconius numata, several wing colour patterns are co-existing and these patterns exhibit high resemblance with other unpalatable species. Colour patterns thus seem to act as a warning signal of toxicity for predators. In H. numata, this protective mimicry is adapted to the spatial variation in communities of unpalatable butterflies, leading to a stable polymorphism of wing colour patterns due to selection/migration equilibrium. These complex wing colour patterns are mainly controlled by a single locus, the supergene *P*, which contains about 18 co-segregating genes. Dominance relationships among the haplotypes at the supergene *P* are predicted to be under high selective constraint due to an increased predation risk for non-mimetic intermediate heterozygotes. Using an original morphological approach based on automatic detection of pattern variation, we quantify the coefficient of dominance between haplotypes at the supergene P. The study of controlled crosses between sympatric and allopatric morphs suggests a complete dominance among haplotypes occurring within populations whereas mosaic of dominance is mainly observed in allopatric haplotypes. This highlights the important role of dominance in the polymorphism of distinct wing colour patterns involved in mimicry relationships and allow us to open research on the possible mechanisms of dominance at the supergene *P*.

D21SY21PS0796

FLOWER COLOUR MORPHS OF IRIS PUMILA DIFFER IN THE AMOUNTS OF HSP90 AND PHENOLIC COMPOUNDS

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Natural populations of the dwarf bearded iris, Iris pumila, display a striking flower-colour polymorphism. It was noted that the combination of fluctuating temperatures and the varied attractiveness of diverse colour morphs promotes a stable coexistence of multiple colour variants in a population. We have quantified the amounts of Hsp90 and the antioxidants, anthocyanins and total phenolics, in *I. pumila* flowers. These molecules impact abiotic stress tolerance, ultimately influencing the fitness of individual plants. A total of 100 clones that were raised in a common garden and assessed to different colour classes (dark violet, violet, light violet, dark blue, light blue, yellow/white) were examined. The amounts of two Hsp90 forms, inducible (Hsp90a) and constitutively expressed (Hsp90b) proteins were lowest in yellow and white flowers as compared to other colour classes. In blue flowers, the concentration of Hsp90a was observed to decrease gradually when proceeding from light blue to dark blue variants, whereas an inverse trend was observed in violet-coloured flowers. The concentration of anthocyanins was notably low in white/yellow flowers and in the blue and violet colour classes it progressively increased from light to dark floral morphs. The amounts of total phenolics were highest in the white and yellow colour morphs; they were relatively high in all of the blue variants, and gradually increase from light to dark colour morphs in the violet class. These results suggest that each *I. pumila* colour genotype is responsible for the production of unique amounts of Hsp90 and phenolics that protect cellular homeostasis under fluctuating temperature conditions within populations.

D21SY21PS0859

GENOMIC INSTABILITY IN THE LOCUS RESPONSIBLE FOR A CONSPICUOUS POLYCHROMATISM IN NICARAGUAN MIDAS CICHLID FISH

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Body coloration is polymorphic in many species. It is subject to both natural selection (e.g. cryptic coloration) and sexual selection (e.g. male nuptial coloration). Two extraordinary features of Cichlids are their enormous diversity in coloration and their rapid rates of diversification. Sexual selection on coloration has been proposed to be one of the major forces driving their explosive speciation rates, which makes them an ideal system to study the effects of body coloration on speciation processes. Most of the Nicaraguan lakes are inhabited by two color morphs of Midas cichlids: a barred dark morph, which represents the majority of individuals (~90%), and a gold morph (7 - 10%). All individuals have the normal phenotype at early stages of their life but some lose their dark coloration and become gold. Gold and normal fish mate assortatively and genetic divergence of neutral markers occurs between the two morphs. All of which suggests a role of this trait in the process of incipient speciation. In this study, we characterize the architecture of the genomic region that harbors the causal gold polymorphism. The interval was reduced to approximately 60 kb and it, as well as the flanking regions, shows strong evidence of genomic instability. This includes several indels of various sizes, gene duplications and chromosomal rearrangements. Lineage-specific, tandemly duplicated genes have been identified, some of which show signs of pseudogenization and selection. Comparative genomic analysis of the region indicates the presence of cichlid-specific and perhaps even Midas-specific chromosomal rearrangements. Regions of genomic instability have been proposed to have an impact on speciation due to altered recombination and mutation rates. The present study illustrates the architecture of an unstable genomic region that might underlie incipient sympatric speciation in Nicaraguan Midas cichlids.

D21SY21PS0882

DIFFERENTIAL EFFECTS OF PIGMENTARY AND NON-PIGMENTARY ANTIOXIDANTS ON GROWTH, PLUMAGE COLORATION AND RESISTANCE TO OXIDATIVE STRESS IN WILD GREAT TITS

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Carotenoid-based colorations are thought to be honest signals of individual quality but the mechanism underlying their expression is still not clear. Since carotenoids act both as antioxidants and immunostimulants, it has been suggested that carotenoid-based coloration can signal an individual's ability to resist oxidative stress and/or to mount an immune response. However, the antioxidant role of carotenoids in vivo has recently been debated. The "protection hypothesis" holds that carotenoids, which are minor antioxidants and are bleached by reactive oxygen species, can be used as signals to indicate the availability of non-pigmentary antioxidants (e.g. vitamins) that protect them from oxidation. Here, we evaluated this hypothesis by assessing the interactive effects of carotenoids and vitamins on plumage coloration, oxidative stress, growth and fledging success in nestling great tits. We supplemented nestlings with carotenoids (lutein and zeaxanthin) and/or vitamins (E and C) in a 2x2 full-factorial design, and subsequently measured plumage reflectance, antioxidant capacity, oxidative damage and body condition. Vitamins enhanced the expression of the carotenoid-based plumage coloration during the breeding season and improved antioxidant capacity. They did not influence oxidative damage, probably because supplied nestlings invested more in growth, which is a major cause of free-radical production, rather than in reducing oxidative damage. Moreover, vitamin-treated nestlings had a higher probability of fledging. In contrast, carotenoids did not influence any of these traits and did not show any synergistic effect when supplemented together with vitamins. Our results support the "protection hypothesis" and hence the idea that carotenoids are minor antioxidants in vivo. Furthermore, we could show the importance of antioxidants during growth, supporting the idea that oxidative stress may play a central role in life-history trade-offs.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS0903

EXPRESSION LEVELS OF GENES BELONGING TO THE MELANOCORTIN SYSTEM ARE ASSOCIATED WITH MELANIN-BASED COLORATION IN TWO COLOUR POLYMORPHIC OWL SPECIES

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The adaptive function of colour polymorphism is a long-standing debate, principally because of limited knowledge of the genetic mechanism underlying morph production. A recent genetic model suggested that the melanocortin system could account for covariations between melanin-based colour morphs, behaviour, morphology and physiology. This genetic system may therefore account for the observed morph-specific life history strategies. In two owl species we explored whether the expression levels of genes belonging to the melanocortin system (MC1R, POMC, PC1, PC2 and the antagonist ASIP) as well as 15 other melanogenic genes are associated with melanin-based coloration. We considered the tawny owl (Strix aluco) because individuals vary continuously from light to dark reddish. We measured gene expression in feather follicles collected in nestlings at the time of melanin production. Our results are consistent with a key role of the melanocortin system on the expression of colour morphs. We indeed found that the expression levels of convertases (that process melanocortin hormones) covary with melanin-based coloration, an effect that strongly depends on genetic polymorphism at the melanocortin-1-receptor (MC1R). We conclude that the melanocortin system may explain why dark and light melanic morphs adopt alternative life history strategies and differentially cope with stressful factors.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS0912

BEING COMPLEX CAN BE SAFE: TESTING PREDATOR AVOIDANCE OF VARYING APOSEMATIC SIGNALS IN A NOVEL ENVIRONMENT

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Conspicuous colour patterns may function as interspecific signals in the context of predation by warning predators about prey unprofitability (aposematism). This strategy relies on the ability of predators to learn the association between colouration and unprofitability, and the subsequent avoidance of the unprofitable prey. Frequency-dependent selection is expected to favour uniformity and act against variability in aposematic signals. However, variation in aposematic species occurs in many taxa suggesting that signal variation may serve other purposes or be under variable selective pressures. Although the fundamental assumptions of aposematism have been well supported by mathematical models and experiments in controlled laboratory setups, their implications in the natural environment of both predators and prey have been until recently greatly overlooked. Predators are supposed to learn simpler patterns easier. Because variation in aposematic signals may imply morph-specific attack rates, aposematism as an anti-predator strategy could be overall less effective for individuals with complex colour patterns, especially when exposed to naïve predators or when invading novel environments, unless there were associated differences in detectability. We tested that hypothesis using wax models of the polymorphic, aposematic poison frog Dendrobates tinctorius placed in the wild, in a site where the actual frogs do not occur. We found that over time aposematic prey get less attacks than cryptic prey, but there were no differences in the attack rate between simple and complex morphs. However, complex morphs seemed to be more difficult to detect than simple ones. We suggest that wild predators are able to generalise aposematic colour patterns. Complex patterns may compensate being difficult to learn by being less detectable, which may contribute to the maintenance of the great intra-populational variation in colour patterns in this species.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS1212

MAPPING OF THE GUPPY PIGMENTATION GENE BLOND USING RAD-SEQ

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The guppy (*Poecilia reticulata*) is a long-standing model for the study of sex-linked inheritance of male ornamental traits. The species also offers excellent opportunities to observe 'evolution in action,' as its populations adapt rapidly to changing predator regimes. While numerous studies have investigated the delicate interplay between natural and sexual selection shaping male coloration in the wild, surprisingly little is known about the genes and developmental pathways underlying guppy coloration. We have shown previously that spontaneous mutations in *kita* and *colony-stimulating factor 1 receptor a* (*csf1ra*) significantly change the normal camouflage as well as the variable male ornaments of the guppy. Another spontaneous guppy mutant, *blond*, greatly reduces all black pigmentation and thereby dramatically changes male color patterns. To map this pigmentation gene, we used next-generation sequencing followed by a quantitative trait locus (QTL) analysis. A total of 141 F2 individuals from a cross between a wild-type female and a blond male were sequenced by representational analysis using the restriction-site associated DNA tag method (RAD-seq). RAD-seq reads were mapped to a draft version of the guppy genome and single-nucleotide polymorphisms (SNPs) between the individuals were identified. The resulting ~7,000 high quality SNPs were used to narrow down a major QTL spanning 118 kb on autosomal linkage group 2. The target region contains three genes, of which adenylate cyclase 5 (adcy5) is a promising candidate gene as preliminary analysis suggests that it contains a frameshift mutation in blond guppies. Our study demonstrates that RAD-seq is a powerful tool to identify genes of interest even in organisms for which only limited genomic information is available.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS1270

MAPPING THE CEPAEA NEMORALIS SHELL COLOUR AND BANDING SUPERGENE USING RAD-SEQ

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The classic shell colour and banding polymorphism of the land snail *Cepaea nemoralis* played a crucial role in establishing the importance of natural selection in maintaining morphological variation. *Cepaea* is also a pre-eminent model for ecological genetics because the outward colour and banding phenotype is entirely genetically determined, primarily by a 'supergene' of at least five loci. Unfortunately, progress in understanding the evolution and maintenance of the *Cepaea* polymorphism stalled, partly because of a lack of genetic markers. However, high-throughput genotyping-by-sequencing technologies, such as Restriction Site Associated DNA sequencing (RAD-Seq), now enable systems with few existing genetic resources to be researched from new angles and in far greater detail than ever before. Consequently, with an ultimate view to identify and dissect the individual components of the Cepaea supergene, and to understand their evolution, we used RAD-Seq to construct a local map of markers linked to the shell ground colour (*C*) and banding presence (*B*) loci of the supergene. We have identified and validated 11 linked markers, with the closest being within ~0.6 cM (1 recombinant out of 168) of the *C*-*B* supergene linkage group, with the combined loci together forming a 35.8 cM linkage map of markers that flank both sides of the Cepaea C-B supergene. This initial set of linked markers should form the foundations of future work to re-establish *Cepaea* as a prominent model of molecular ecology.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS1290

THE ROLE OF MELANIN PATHWAY GENES IN NATURAL VARIATION OF PIGMENTATION TRAITS IN DROSOPHILA MELANOGASTER

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Research on insect pigmentation has been informative in determining how evolutionary mechanisms can produce high levels of animal diversity. Scientists are only beginning to understand how pigmentation genes may contribute to natural diversity in pigmentation traits. Identifying these genes, their functional roles, and impact on population-level processes will elucidate how genetic components may contribute to the natural variation, adaptation, and evolution of a species. This study attempts to do this by closely examining the candidate pigmentation genes *pale* and *ebony* in the fruit fly species, Drosophila melanogaster. I conducted a SNP assocation study with pigmentation traits in isogenic flies derived from a population in Raleigh, NC. SNPs within coding and non-coding regions of pale and ebony were significantly associated with phenotypic variation. This suggests a putative role of these genes and specific genic regions in pigmentation variation. Phenotypic data was also collected in individuals derived from five geographically distinct populations in the Eastern United States. Significant variation was identified among lines and populations, suggesting geographic patterns in these traits and a possible contribution of pigmentation to adaptation. Phenotype data from these populations will be used in conjunction with sequence data to determine if these genes are contributing to variation in other populations, whether there are informative geographical patterns in SNP or haplotype frequency, and if there are signatures of natural selection. My work will allow me to determine the extent that *ebony* and *pale* are involved in natural variation in pigmentation in D. *melanogaster* and uncover possible underlying evolutionary mechanisms. Additionally, this knowledge can be applied to research on the evolution of other pigmentation genes and mechanisms responsible for species diversification and adaptation.

21. The Evolution and Maintenance of Heritable Colour Polymorphisms: from Ecology to Genomes

D21SY21PS1366

ROLE OF FREQUENCY DEPENDENT SELECTION ON THE MAINTENANCE OF POLYMORPHISM IN APOSEMATIC ORGANISMS

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The maintenance of trait polymorphisms in aposematic organisms is an evolutionary puzzle. The advantage of predator learning should drive warning signals of prey towards monomorphism, yet there are several examples of polymorphic aposematic species. Solving the riddle of polymorphism in such challenging cases would therefore represent an important contribution to our understanding of polymorphism maintenance in nature. Here, we examine the role of frequency dependent selection (FDS) on the maintenance of color polymorphism in an aposematic species. While models of frequency dependent selection are common, empirical evaluations are rare. The aposematic wood tiger moth (Parasemia plantaginis) presents two distinct male color morphs (yellow and white). Some populations have an admixture of both morphs whereas a few populations present only one of them. Research has previously shown that the more conspicuous yellow morph has a survival advantage under predation, which increases with morph frequency (i.e. positive FDS). This advantage and positive FDS should lead the yellow morph to fixation, yet this is not the case. We hypothesized that counteracting reproductive FDS (negative or positive) could maintain the polymorphism. First, we tested for reproductive FDS in the species using semi-natural enclosures set with three different frequencies of male color morphs (control, white-biased, and yellow-biased) to examine if the rare or common morph has a mating advantage. The experiment showed positive FDS in mating success for both morphs. Second, we formulated a theoretical model to test whether these results, and previous results on survival, can explain the maintenance of the observed polymorphism. The model showed that our patterns of positive frequency dependence can lead to polymorphism if considered in a spatial context, where variation in interacting ecological factors (e.g. predation), combined with a very small amount of gene flow can maintain the polymorphism.

Symposium

22. The Evolution and Genetics of Drug Resistance

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3, 4

Organisers: Pleuni Pennings, Gabriel G. Perron, Sarah Cobey and R.

Fredrik Inglis

Invited speakers: Craig MacLean

Description:

The evolution of drug resistance in pathogenic microorganisms is one of the most important challenges facing evolutionary biologists. Evolutionary studies of drug resistance can aid the development of effective clinical strategies. At the same time, such studies help further our general understanding of evolutionary biology. Our symposium provides a venue to discuss experimental and theoretical studies that improve basic understanding and/or inform clinical practice.

D20SY22IT10:30R6

THE EVOLUTIONARY CAUSES AND CONSEQUENCES OF DIVERGENCE IN ANTIBIOTIC RESISTANCE BETWEEN BACTERIAL SPECIES: INSIGHTS FROM COMPARATIVE AND EXPERIMENTAL STUDIES

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This talk will address two unresolved problems: i) Why do species of bacteria differ in their intrinsic resistance to low levels of antibiotics that are found in natural environments? And, ii) How does variation between species of bacteria impact their ability to evolve resistance to high levels of antibiotics that are found in clinical environments? To address these questions, we study antibiotic resistance in bacteria from the genus Pseudomonas: this genus of bacteria shows exceptional levels of phenotypic and genetic diversity, and antibiotic resistance in P.aeruginosa has emerged an important clinical problem. First, I will show how integrating phenotypic and functional genetic measures of resistance into a comparative framework can be used to infer historical patterns of selection on antibiotic resistance and identify the genetic mechanisms that underlie variation in resistance levels between species of bacteria. This comparative work shows that selection drives divergence in resistance among species, and that the underlying genetic mechanism driving this divergence is changes in the number of antibiotic efflux pumps. Second, I will present the results of experiments that explore the ability of different species of Pseudomonas to adapt to a common selective pressure, in the form of a lethal dose of the antibiotic rifampicin. These experiments show that different species vary substantially in their ability to evolve rifampicin resistance, and that this divergence is driven by variation in the mutation spectrum and epistasis for fitness across species. Importantly, we find no evidence of a connection between intrinsic resistance and the ability to evolve high levels of antibiotic resistance. In a broader evolutionary framework, this talk will provide novel insights into how and why species show convergent and divergent adaptations to common selective pressures at a molecular and phenotypic level.

D20SY22RT11:18R6

THE ROLE OF ANTIRETROVIRAL DYNAMICS IN THE EVOLUTION OF DRUG RESISTANCE IN HIV

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Despite the high inhibition of viral replication achieved by current anti-HIV drugs, many patients fail treatment, often with emergence of drug-resistant virus. Clinical observations show that the relationship between adherence and likelihood of resistance differs dramatically among drug classes. We developed an evolutionary model that explains these observations and predicts treatment outcomes. Our model incorporates drug pharmacokinetics and pharmacodynamics, fitness differences between susceptible and resistant strains, mutations and patient adherence measures. We show that antiviral activity falls quickly for drugs with sharp dose-response curves and short half-lives, such as boosted protease inhibitors, limiting the time during which resistance can be selected for. We find that poor adherence to such drugs causes treatment failure via growth of susceptible virus, explaining puzzling clinical observations. We examine both monotherapy and combination therapy, demonstrating how the concept of the 'mutant selection window' can help explain HIV resistance. Furthermore, our model predicts that certain single-pill combination therapies can prevent resistance, even in the case of imperfect adherence and "drug holidays. We use our results to prioritize a wide range of dual- and triple- therapies based on expected clinical outcomes. Our approach represents a first step for simulating clinical trials of untested anti-HIV regimens and may help in the selection of new drug regimens for investigation. More generally, we show how fluctuating drug concentrations exacerbate the problem of resistance compared to constant doses with the same time averaged concentration or inhibition level, especially when multiple mutations are needed for resistance.

D20SY22RT11:42R6

EVOLUTIONARY GENOMICS OF EPIDEMIC AND NON-EPIDEMIC STRAINS OF PSEUDOMONAS AERUGINOSA

<u>Jeremy R Dettman</u>¹, Nicolas Rodrigue², Rees Kassen¹ ¹University of Ottawa, Canada ²Agriculture and Agri-Food Canada, Canada jdettman@uottawa.ca

Pseudomonas aeruginosa is an opportunistic pathogen of humans and is the most common bacterial species isolated from the respiratory tracts of adult patients with cystic fibrosis (CF). Chronic infection of the CF lung can lead to decades of direct interaction between the host and resident *P. aeruginosa* population. Longitudinal studies have documented the patterns of adaptation to the CF lung, and evidence to date suggests that a large number of genes are targets for mutation, but most are mutated in only a small fraction of infections. A more comprehensive view therefore requires the comparison of a larger sample of diverse clinical isolates. To this end, we obtained whole genome sequence data from a collection of *P. aeruginosa* isolated from the airways of CF patients in order to investigate general patterns of adaptation associated with chronic infection. We also focus attention on a transmissible, epidemic strain that was recently reported within North America. We present multiple lines of evidence that the history of selection imposed by the CF lung environment has a major influence on genomic evolution and the genetic characteristics of isolates causing contemporary infection. We identify candidate genes and important functional pathways, and find that the presence of oxidative stressors and antibiotics appear to be key factors that have driven the adaptive evolution of this pathogen within the host.

D20SY22RT14:00R6

SELECTION, CHARACTERIZATION AND THE WITHIN-HOST ECOLOGY OF DRUG RESISTANT MALARIA PARASITES

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The evolution of drug resistance is a key challenge for our ability to treat and control infections. For malaria parasites artemisinin based drugs are the front line weapons in the fight against disease, but reports from the field of slower clearance rates, are causing concern that the useful life-span of these drugs may be limited. Understanding whether slower clearance rates represent true resistance, and predicting the speed at which this could spread in the field, is challenging due to host effects on clearance rates and the lack of molecular markers. Here we show that *Plasmodium chabaudi* malaria parasites selected for Artesunate resistance evolve slower clearance rates, similar to those seen in the field, extremely rapidly. These slower clearance rates provide a fitness advantage to parasites exposed to drugs within single infections where overall parasite density, relapse after treatment and transmission potential is higher. Within mixed infections, the fitness advantage is even stronger, as selected parasites experience release from competition with susceptible strains, resulting in higher total parasite densities and greater transmission potential. Additionally, the magnitude of competitive release within mixed infections is dependent on the strength of drug treatment. More aggressive treatment results in a stronger selective advantage to resistant parasites, despite not providing an additional health benefit to hosts when compared to treatment with lower drug doses. Our results show that slower clearance rates can rapidly evolve and provide a strong fitness advantage both in single and mixed strain infections, suggesting this phenotype has the potential to spread within parasite populations. Furthermore, drug dose dramatically influences fitness and transmission potential of resistant parasites, suggesting careful consideration should be given to selecting optimal treatment regimes.

D20SY22RT14:24R6

DEADLY ANTIBIOTICS AND EVOLUTIONARY RESCUE VIA HORIZONTAL GENE TRANSFER

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Evolution has caused one of the most concrete problems in the modern world by selecting for drug resistant bacteria. When bacteria are exposed to lethal levels of antibiotics, it has been assumed that there needs to be a pre-existing mutation for resistance within the population in order for the bacterial strain to survive the drug treatment. However and given that majority of resistance genes are carried by mobile genetic elements such as conjugative plasmids that can move between bacterial cells, it is possible that the resistance gene pre-exist in another bacteria may survive lethal antibiotic selection by acquiring resistance genes horizontally, and, if so, could this process be somehow prevented. Indeed and contradictory to previous paradigm, we demonstrate that the resistance genes. Moreover, we show that viruses specifically infecting bacteria with mobile elements can both prevent the spread of resistances to other bacteria and to lead to the loss of resistance conferring elements from the population. Altogether these results suggest that the composition of bacterial community along with their viruses can play a defining role in the evolution antibiotic resistances.

D20SY22RT14:48R6

ANTIBIOTIC RESISTANCE AND STRESS IN THE LIGHT OF FISHER'S MODEL

<u>Ana Sousa</u>¹, Sandra Trindade¹, Isabel Gordo¹ ¹Evolutionary Biology, Instituto Gulbenkian de Ciencia, Portugal amsousa@igc.gulbenkian.pt

The role of mutations in evolution depends upon the distribution of their effects on fitness. This distribution is likely to depend on the environment. Indeed genotype-by-environment interactions are key for the process of local adaptation and ecological specialization. An important trait in bacterial evolution is antibiotic resistance, which presents a clear case of change in the direction of selection between environments with and without antibiotics. Here, we study the distribution of fitness effects of mutations, conferring antibiotic resistance to Escherichia coli, in benign and stressful environments without drugs.We interpret the distributions in the light of a fitness landscape model that assumes a single fitness peak. We find that mutation effects (s) arewell described by a shifted gamma distribution, with a shift parameter that reflects the distance to the fitness peak and varies across environments. Consistent with the theoretical predictions of Fisher's geometrical model, with a Gaussian relationship between phenotype and fitness, we find that the main effect of stress is to increase the variance in s. Our findings are in agreement with the results of a recent meta-analysis, which suggest that a simple fitness landscape model may capture the variation of mutation effects across species and environments.

D20SY22RT15:12R6

EFFECT OF PHAGES ON THE COST OF ANTIBIOTIC RESISTANCE

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Experimental studies of antibiotic resistance evolution typically employ a single bacterial species in pure culture. However, outside the laboratory, parasitism of bacteria by viruses (phages) is ubiquitous. I used experimental populations of *Escherichia coli* to show that phage parasitism altered the outcomes of competitions between antibiotic-resistant and sensitive genotypes. Phages caused rapid selective sweeps, associated with the emergence of phage-resistant mutants, and this was approximately equally likely to favour antibiotic-resistant or sensitive genotypes when their initial frequencies were similar. An elevated mutation rate was advantageous in the presence of phages, suggesting that a given antibiotic-resistant or sensitive genotypes. Given their abundance in nature, phages probably modify selection on other traits in many different scenarios.

D20SY22RT15:45R6

ANTIBIOTIC RESISTANCE AMONG THE KILLERS

<u>Daniel E Rozen</u>¹, Ard Jan Grimbergen¹ ¹*microbial biotechnology, institute of biology, leiden university, Netherlands* d.e.rozen@biology.leidenuniv.nl

The discovery and development of antibiotics as part of the medical arsenal is one of the great triumphs in the effort to eradicate bacterial diseases. For more than 70 years these microbial products have worked with remarkable success, transforming the medical landscape and dramatically improving human health. However, despite their tremendous relevance for humans, there is a surprising lack of understanding of the role of antibiotics in nature for the organisms that produce them. Why are antibiotics produced and why do strains in nature evolve to resist them. Here we test the idea that antibiotic-mediated interference competition between coexisting bacterial species in soil drives reciprocal coevolutionary changes as strains evolve novel mechanisms of killing and resistance. Focusing on the prolific antibiotic producing genus, Streptomyces, and their coexisting competitors, we first characterize interaction networks between coexisting Streptomycetes and then ask whether antibiotic resistance shows evidence of local adaptation. Second, we are investigating the costs and in situ fitness of naturally resistant strains of Streptomyces in soil at different antibiotic concentrations and during co-cultivation with Streptomycetes that are natural antibiotic producers. Our work examines the natural context and population dynamics of resistance evolution, thereby providing insight into processes occurring in the clinical environment.

D20SY22RT16:09R6

DIRECT BENEFIT OF THE SOS RESPONSE VANISHES AFTER LONG TERM EVOLUTION IN P. AERUGINOSA

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Exposure to stressors, such as antibiotics, induces the expression of response pathways that could provide bacteria with a direct fitness benefit, stemming from increased protection against stress, and an indirect fitness benefit, stemming from increased evolvability due to elevated mutagenesis. We have guite a good understanding of the molecular mechanisms of stress response pathways, but quantitative measurements of the direct and indirect fitness benefits and costs associated with stress response pathway expression are lacking. By measuring the competitive fitness of wildtype and LexA mutant (SOS response blocked) strains of *P. aeruginosa* we show that the SOS response provides with a direct benefit across a range of sub-lethal concentrations of the antibiotic ciprofloxacin. Interestingly, in the absence of the antibiotic, the LexA mutation confers an advantage in the competitive ability and demonstrates the existence of a cost for the cells owning this complex pathway. We also analyse important parameters like cell viability, cell size and growth rate to explain the higher fitness of wildtype during selection with antibiotic. However, we find that the SOS pathway produces no indirect fitness benefit, as demonstrated by the fact that the LexA mutant shows a comparable ability to adapt to ciprofloxacin over a 600 generation selection experiment. Our study clearly demonstrates that natural selection favours the evolution of stress-induced mutagenesis pathways as a result of direct selection for increased stress tolerance and not indirect selection to favour evolvability. Altogether, we contribute with experimental evidence crucial to disentangle if bacterial stress responses play a role in accelerating adaptation to stressors such as antibiotics.

D20SY22RT16:33R6

WHY ANTI-MALARIAL DRUG RESISTANCE IS SPREADING FASTER IN PLASMODIUM FALCIPARUM THAN IN PLASMODIUM VIVAX

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Affiliation: Full Professor, University of applied Sciences Mittweida, Germany Abstract: Drug treatments - in the absence of vaccines - are a major component of malaria control program. The two human malaria agents, P. falciparum and P. vivax, causing the most morbidity and mortality have distinct characteristics that affect their treatments. Considering these species-specific differences is crucial for control and elimination efforts. Particularly, the rapid evolution of drug-resistant P. falciparum is currently a source of concern for global health. A complex compound of particular steps (fitness components) in the malaria life cycle determines evolutionary fitness of drug-resistanceconferring mutations. Differences in fitness components affect how selection imposed by interventions, e.g. drug treatments, differentially acts on each Plasmodium species. We formulate a populationgenetic model that explains how different fitness components affect selection for drug resistance, focusing on: (i) characteristics of sexual parasite life stages responsible for host-to vector transmission; and (ii) presence of dormant liver-stage parasites. The model explains why drug-resistance is evolving much faster in P. falciparum than in P. vivax. The model's data-driven nature (i.e., parameters can be estimated from molecular data) has the potential to facilitate new experimental designs to better understand drug-resistance evolution in terms of clinically measurable quantities. A successful malaria control/elimination program should incorporate how differences in fitness components across malaria species affect adaptive evolution (e.g. the emergence of drug resistance). Reference: Schneider, KA, Escalante, AA (2013). Fitness components and natural selection: why are there different patterns on the emergence of drug resistance in P. falciparum and P. vivax? Malaria J. 12: 15.

D20SY22RT16:57R6

MODELLING ANTIBIOTIC RESISTANCE AND PLASMIDS

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The spread of antibiotic resistance in bacteria is a major public health problem, threatening our ability to treat bacterial infections successfully. Plasmids, extra-chromosomal pieces of DNA capable of horizontal transfer, are often implicated in the process of resistance acquisition, and potentially allow for the transfer of resistance genes between species and between genera. Some plasmids carry multiple resistance genes, simultaneously counteracting several antibiotics, and in extreme cases acquisition of a plasmid by a pathogen can confer resistance to virtually all antibiotics at a single stroke. The conditions favouring resistance genes to be located on plasmids rather than on chromosomes are therefore potentially of great importance. Mathematical models are an excellent technique for investigating this topic, because they allow for simultaneous consideration of selection at three levels: genes, plasmids, and bacteria. We use models to consider whether plasmids are a favourable location for antibiotic resistance genes, and how spatially- or temporally-varying selection regimes can affect this. Answers to these questions provide a more complete understanding of the processes underlying the evolution of antibiotic resistance genes on plasmids, and could consequently be of great value in preventing the spread of resistance.

D20SY22RT17:45R6

EXPLORING THE EVOLVABILITY OF AN ANTIBIOTIC RESISTANCE ENZYME

<u>Arjan DeVisser</u>¹, Martijn Schenk¹, Merijn Salverda¹, Ivan Szendro², Joachim Krug² ¹Laboratory of Genetics, Wageningen University, Netherlands ²Institute for Theoretical Physics, University of Cologne, Germany arjan.devisser@wur.nl

For a quantitative understanding of the process of adaptation, we need to understand its 'raw material', that is the properties of beneficial mutations. In my talk, I will focus on two properties in particular, the frequency distribution of fitness effects of beneficial mutations and their epistatic interactions, and how these determine the pathway and outcome of evolution. In the experiments that I will present, we study the in vitro evolution of the enzyme TEM-1 beta-lactamase, a notorious determinant of antibiotic resistance in bacteria. The first two studies are systematic investigations of the short-term evolvability of the enzyme, including the number and effects of beneficial mutations and their epistatic interactions. The last two studies address the role of the structure of the fitness landscape (caused by epistasis) and population size on long-term evolvability. Surprisingly, we find that small populations sometimes reach higher resistance than large populations, showing the important role of chance events for long-term adaptation.

D20SY22RT18:09R6

THE ROLE OF DRUG PENETRATION IN THE EVOLUTION OF MULTI-DRUG RESISTANCE

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The emergence of resistance in combination therapy can be facilitated by treatment regimes where only one drug is active at certain periods of time, resulting in *temporal* effective monotherapy (e.g. structured treatment interruptions). This occurs because resistance mutations can arise in a stepwise instead of a concurrent manner when only one drug reaches an effective concentration. We propose that effective monotherapy can also occur at a *spatial* level because of discordant penetration of drugs into compartments of the body. Here, we present a mathematical model for the evolution of drug resistance in combination therapy when there is differential drug penetration. Our model shows that the presence of body compartments where only one drug in a combination regime penetrates to a therapeutic concentration strongly speeds up the evolution of multi-drug resistance. Resistance mutations are acquired in a stepwise manner through colonization of body compartments where the pathogen can survive without being resistant to all the drugs in the regime. These results offer a possible explanation for the evolution of single-drug resistance in the course of combination therapy. We compare our model results to clinical trials for antiretrovirals and antibiotics to identify cases where stepwise acquisition of mutations can be facilitated by differential drug penetration. Overall, our results suggest that the use of drugs with similar penetration profiles in combination therapy may prevent the evolution of multi-drug resistance.

D20SY22RT18:33R6

EXTRACELLULAR QUENCHING OF BACTERIAL PUBLIC GOODS AS AN 'EVOLUTION-PROOF' ANTI-VIRULENCE THERAPY: A CASE STUDY WITH PSEUDOMONAS AERUGINOSA

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Many bacterial exoproducts yield population-level benefits. Such 'public goods' (PG) include key virulence factors, and therapies designed to block their production are attracting increasing attention nowadays. The disruption of cell-to-cell communication (quorum sensing, QS), is considered especially promising because it could block production of multiple exoproducts, yet should prompt weaker selection for resistance than conventional antimicrobials. However, initial enthusiasm for this approach ('quorum quenching', QQ) has been tempered by claims that resistance in fact evolves readily, for example by improving pumps to eject QQ compounds from cells. Here, we focus on a different strategy, in which PGs (siderophores) are quenched outside producer cells. We show that adding gallium (Ga) to iron-limited Pseudomonas aeruginosa cultures suppresses growth in a dosedependent manner by (a) deactivating siderophores and thereby choking the supply of iron, and (b) inducing costly production of further siderophores. In experimental infections of moth larvae (Galleria mellonella), Ga suppressed bacterial growth and extended larval survival. Crucially, moderate levels of Ga reduced virulence below those of infections with siderophore-defective mutant strains, which suggests that Ga also induces siderophore overproduction in vivo, imposing extra metabolic burden on bacteria without generating benefits. We argue that strategies that quenching secreted PGs extracellularly should be more effective than those that inhibit synthesis in the cell, since PG production costs remain or even increase. With Ga-mediated PG-quenching, resistance is particularly unlikely to evolve because (a) extracellular quenching is impervious to typical within-cell resistance traits; (b) avoiding siderophore production is maladaptive (non-producing 'cheats' could not spread in our experiments); and (c) evolving siderophores with reduced susceptibility to Ga appears to be biochemically unfeasible.

D20SY22RT18:57R6

MULTILEVEL EVOLUTIONARY CHANGES AFTER HORIZONTAL GENE TRANSFER OF AN ANTIBIOTIC RESISTANCE GENE

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Horizontal gene transfer is a powerful mechanism by which antibiotic resistance spread among species. However, the maintenance of a horizontally transferred antibiotic resistance gene is not granted, because of its low level of adaptation to the receiving organism. In particular, transferred genes usually have codon preferences that differ from the ones of the receiving genome. This leads to translation errors, low translation rates and finally to low activity of the transferred gene and energetic costs for the receiving cell. It is theoretically predicted that a deoptimized codon usage gene will undergo an amelioration process - i.e. its sequence will evolve towards a codon usage similar to the host one - and a fine tuning of its expression and of the expression of interacting genes. To test these predictions, three synonymous versions of the Chloramphenicol Acetyl Transferase with different codon preferences were designed and transfected in *Escherichia coli*. After verifying that unadapted codon usage induced a fitness cost, we experimentally evolved these populations for 1000 generations. At the phenotypic level, the cost of codon usage deadaptation was totally compensated. However, at the genotypic level, no amelioration process was observed but various genetic changes occurred ranging from mutations in the promoter to gene loss to plasmid copy number reduction and to genetic changes in the bacteria chromosome. At the proteome level, the presence of a codon usage biased gene and the posterior evolution affected the expression of a high number of proteins. Our results suggest that the presence of horizontally transferred gene with a different codon usage induces selection pressures leading to a refactorization of the bacterial functioning. Horizontal transfer of antibiotic resistance gene thus reveals itself, not only as a spreading highway for these genes but also as a powerful mechanism pushing bacteria to explore new ways of functioning.

POSTERS

AN ADAPTIVE EVOLUTION OF HELICOBACTER PYLORI (H.PYLORI): WHAT IS A ROLE CAG-A LOSS IN THE ERA OF ANTIBACTERIAL TREATMENT?

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A resistance to antibacterial drugs is a classic demonstration of natural selection in microorganisms during the period of host-bacterial interaction and high intensity of antibiotic exposure. The aim of the study was to determine the evolutionary role of the CagA pathogenicity island (PAI) presence in the outcome of eradication treatment of H.pylori. Sixty-seven pediatric patients with upper dyspepsia (mean age 13.7 years, range 5 to 17 years, male/female 24/43) undergoing endoscopy were investigated for H.pylori presence. Gastric antral biopsy specimens were taken for histology and/or culture and one biopsy fragment was used for CagA PCR determination. H.pylori+ (positive) patients were treated with a two-week course of amoxycilline (50 mg/kg/day), bismuth subcitrate (8 mg/kg/day), nifuratel (30 mg/kg/day) plus omeprazole (1 mg/kg, once daily). The criterion for eradication of H.pylori was the negative breath ammonia test. Forty one of 67 children (61.2%) were H.pylori+. Nineteen of 41 strains (46.3%) were CagA+ (group I) and 22 were CagA- (negative) (group II) (53.7%). H.pylori was eradicated in 33 patients (80.4%). Among the patients with successful eradication 19 children were CagA+, 14 - were CagA–. Thus, significantly more patients in group II than in group I had evidence of ongoing H.pylori infection (63.6% (14/22) versus 94.7% (18/19); x2=4.08, p=0.0021). Eight children (19.6%) remained H.pylori+. Obviously, the carriage of CagA-lacking strain was strongly associated with unsuccessful eradication (OR=10.28; 95CI, 1.148 to 92.173). The strains of H.pylori lacking the cag PAI, or parts of it, have lost the island through transformation with an empty site or through deletion mutations. It is possible, that this particular phenomenon, associated with genome plasticity, not just allows the microbe to maintain balanced relationship with the host, but also to survive in antibacterial therapy conditions.

IMPLICATIONS OF STRESS-INDUCED GENETIC VARIATION FOR MINIMIZING MULTIDRUG RESISTANCE IN BACTERIA

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Antibiotic resistance in bacterial infections is a growing threat to public health. Recent evidence shows that when exposed to stressful conditions, some bacteria perform higher rates of mutation and horizontal gene transfer, and thus acquire antibiotic resistance more rapidly. We incorporate this new notion into a mathematical model of ordinary differential equations describing the emergence of antibiotic multi-resistance in a hospital setting. Results: We show that when stress has a considerable effect on genetic variation, the emergence of antibiotic resistance is dramatically affected. In contrast with classical models, a strategy in which patients receive a combination of antibiotics (combining) is expected to facilitate the emergence of multi-resistant bacteria when genetic variation is stress-induced. The preference between a strategy in which one of two effective drugs is assigned randomly to each patient (mixing), and a strategy where only one drug is administered for a specific period of time (cycling) is determined by the resistance acquisition mechanisms. We analyze several features of the mechanisms by which stress affects variation and predict the conditions for success of different antibiotic treatment strategies. These findings should encourage the research of stress-induced genetic variation mechanisms and establish the importance of incorporating data about these mechanisms when considering antibiotic treatment strategies.

D20SY22PS0246

THE STOCHASTIC DYNAMICS OF RESCUE TRAJECTORIES: THE CASE OF THE BACTERIUM P. FLUORESCENS EVOLVING RESISTANCE TO THE ANTIBIOTIC STREPTOMYCIN

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Resistance emergence in microbes is a well-documented example of evolutionary rescue, where beneficial mutations, either present or produced de novo, rescue a population from extinction. Alternative models have been developed in the past two decades emphasizing different key factors affecting the process of emergence. I will present two of these models quickly, then show how empirically measured demographic dynamics under antibiotic stress can provide quantitative insights into the underlying processes. I will present some simple analytical results on the expected patterns (i) for resistance probabilities and (ii) for the stochastic demographic dynamics of rescued populations. I will then compare these expectations with empirical trajectories obtained with the bacterium P.fluorescens (and hopefully E.coli). These rescues take typically one or two steps, and show predictable stochastic behaviour. A substantial portion likely occurred in two steps, from an intermediate mutant with critical growth (close to zero growth rate), and the growth rate of rescues occurred in two steps is typically higher than those occurring in one step. Finally, it proves difficult with these experiments to disentangle rescues from rare standing variants vs. those from a single de novo mutation, although some options to do so will be proposed.

ADAPTATION OF *PSEUDOMONAS AERUGINOSA* TO DIFFERENT ANTIBIOTICS

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Pseudomonas aeruginosa is a widespread Gram-negative bacterium found in water, soil, plants and animals. Its diverse array of virulence factors allows it to establish and proliferate in environments ranging from plant roots to the human respiratory tract. It plays an important role in chronic infections such as in cystic fibrosis (CF), and is the most commonly isolated nosocomial bacterium. Pseudomonads naturally possess multiple response mechanisms against antimicrobial treatments granting it resistance against the most commonly used antibiotics. They range from efflux pump systems and complex genetic regulation to intricate social behaviours like biofilm formation or swarming. However, to date, we lack detailed understanding of the relative importance of each of these mechanisms and their interplay during resistance evolution. Thus, our study uses controlled evolution experiments to evaluate in how far different resistance mechanisms are selectively favored throughout P.aeruginosa adaptation to different types of antibiotics.

UNDERSTANDING THE DYNAMICS OF EVOLUTIONARY RESCUE IN TERMS OF CHANGING ENVIRONMENTS AND REACTION NORMS: AN APPLICATION TO BACTERIAL POPULATIONS

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Populations can go extinct when their environments deteriorate, but evolutionary rescue occurs when a shrinking population adapts to the new environmental conditions. Studies of ecological dynamics of biological populations have ignored their underlying genetic structure. By studying evolution in the context of bacteria, we can link these two aspects and gain a better understanding of the antimicrobial resistance problem. Resistance often comes at a cost to fitness in drug absence. Compensatory mutation can reduce this cost without losing resistance in drug presence. When the compensatory allele is so strong that it has the highest fitness across all environmental conditions (no trade-off), we call it a Superbug. We develop a stochastic simulation model of evolutionary rescue to investigate the roles of 1) different forms of environmental change, 2) the interaction between genotype and environment (reaction norms), and 3) trade-off strength in reaction norms. In particular, we study sensitivity, resistance and compensatory mutation through reaction norms in fluctuating environments (periodically varying drug concentrations). Our model allows us to examine the joint effects of the inter-dose period and drug dosage per single intake. We find that the form of environmental change is critical to evolutionary rescue because it determines whether the population evolves to a fitter phenotype before its fitness falls below a survival threshold. Our results show high enough doses clear the infection effectively, but the period between doses generally has little effect on the outcome. We also find that the Superbug has surprisingly little effect on the probability of extinction and rescue via evolution of compensatory mutation in periodically fluctuating environments.

AZOLE RESISTANCE IN ASPERGILLUS FUMIGATUS: CONDITION-DEPENDENT MUTATIONS FOR RESISTANCE

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Azole resistance in Aspergillus fumigatus: condition-dependent mutations for resistance and compensatory evolution Jianhua Zhang, Sijmen E. Schoustra, Marijke Slakhorst, Bertha Koopmanschap, Alfons J. M. Debets, Paul E. Verweij, Bas J. Zwaan Laboratory of Genetics, Wageningen University; Department of Medical Microbiology, Radboud University, Nijmegen. Abstract: Azole resistance is an emerging problem in the saprophytic mold Aspergillus fumigatus, which is involved in the vast majority of invasive infections in the human lung. Resistance is widespread in the Netherlands with a prevalence of 5.3%. The majority of highly resistant mutants have undergone two mutational events: an insert in the promoter region (tandem repeat TR34) and a point mutation in the structural region of the cyp51 gene (L98H). The occurrence of two mutational events at the same time is highly unlikely to happen by chance. To prevent and stop the spread of azole resistant strains, understanding the condition-dependent occurrence of TR34/L98H resistance and potential compensatory mutations for reducing the cost of resistance in the filamentous fungus A. fumigatus is crucial. We hypothesise that a combination of different modes of reproduction of the fungus facilitates the combination of the cyp51 point mutation with the tandem repeat into one background. In addition, mutagenic and recombinogenic environmental conditions may play an important role in the development of resistance. We present data of our study on the dynamics of TR34/L98H resistance by experimental evolution in the laboratory under various environmental conditions in combination with variation in the life.

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CYP51 PARALOGUES AND AZOLE SENSITIVITY IN RHYNCHOSPORIUM COMMUNE

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The azoles, inhibiting sterol 14- α demethylase encoded by *CYP51*, are widely used both as clinical antifungal drugs and as agricultural fungicides. In contrast to some fungicide classes, no single mutation has arisen that confers broad, qualitative resistance to all azoles, but quantitative sensitivity shifts have arisen through a range of mechanisms. In some cases, sensitivity shifts may be due to enhanced efflux of the drug, or altered sterol metabolism, but other mechanisms involve the fungicide target site: CYP51 mutations confer reduced sensitivity in the clinical pathogen Candida albicans and the wheat pathogen *Mycosphaerella qraminicola*, and *CYP51* over-expression confers reduced sensitivity in the fruit pathogen Venturia inaequalis. Genome sequencing has revealed that some filamentous ascomycetes possess multiple CYP51 paralogues, with CYP51B present in all sequenced species, *CYP51A* in around half, and *CYP51C* only in *Fusarium* species. Presence of *CYP51A* reduces intrinsic azole sensitivity, with *CYP51A* mutations and over-expression resulting in further reductions in sensitivity in clinical isolates of *Aspergillus fumigatus*. However, in the barley pathogen *Rhynchosporium commune*, *CYP51A* is only present in some isolates, and acquired reductions in azole sensitivity in the last fifteen years are linked to the re-emergence of CYP51A in the R. commune population. Historical and functional studies provide evidence for the role of *CYP51A* in reduced azole sensitivity in R. commune.

LOSS OF A COOPERATIVE TRAIT AFFECTS STRAIN COMPETITION IN CYSTIC FIBROSIS LUNG INFECTIONS

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Pseudomonas aeruginosa is an opportunistic human pathogen that is the predominant cause of pulmonary exacerbation in patients with cystic fibrosis (CF), where most individuals succumb to infection at very early stages and subsequently suffer from prolonged chronic infections. *P.aeruginosa* harbours a suite of social and competitive traits. Among these traits is the production of bacteriocins, knows as pyocins. An estimated 90% of *P.aeruginosa* strains produce pyocins and bacteria generally exhibit immunity to their own pyocin production. It has been demonstrated that social traits are lost with increasing colonization time over the course of a pulmonary infection when the bacteria become established, highly adapted and specialized to the CF lung. Pyocins are necessary to colonise the new environment of the CF lung, however, pyocigenicity and its associated competitive ability may be lost once an infection becomes established and chronic, in response to differential selective pressures within the lung. In this study I examine the diversity of pyocin production and resistance profiles of strains isolated from a non-lung environment and strains isolated at progressive stages from a pulmonary CF infection. Reduced pyocigenity and increased susceptibility to pyocins can have important implications in the treatment of *P.aeruginosa* pulmonary infections.

EFFICIENT LINKAGE MAPPING IN SCHISTOSOMA MANSONI USING EXTREME QTL METHODS AND EXOME SEQUENCING

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Classical linkage mapping (LM) can be labor intensive, logistically challenging, expensive and limited in power, because both phenotypes and genotypes must be measured in each of the individual progeny. Extreme QTL (X-QTL, or linkage group selection) methods promise to increase the power and simplify logistics of LM, because many 1000s of progeny can be analysed, and progeny pools rather than individuals are genotyped. In X-QTL analysis large pools of selected and unselected F2 progeny are quantitatively genotyped to identify selected genome regions. We applied this method to examine the basis of oxamniquine (OXA) resistance, a Mendelian trait showing recessive inheritance, in the parasitic fluke Schistosoma mansoni. These parasites have a complex lifecycle utilizing snail intermediate hosts and human definitive hosts (or rodents in laboratory maintenance). We staged a genetic cross between drug sensitive and resistant parasites, then between two F1 progeny, to generate multiple F2 progeny. One group of F2 progeny infecting hamsters were treated with OXA, while a second group was left untreated. We prepared DNA from pooled males and pooled females from treated and untreated F2 populations and then used exome capture to reduce the size of the genome (from 363Mb to 15Mb) for analysis. Exomes from the four groups of pooled progeny (treated and untreated males, treated and untreated females) and the two parents were sequenced to high read depth (>95x) and allele frequencies of 14,489 variants compared between treated and untreated parasites. In both treated male and female pools, we observed dramatic enrichment of alleles (*Z*=11.07, *p*=8.74.10^-29) from the resistant parent in a small region of chr. 6. This region contains SmSULT a gene recently implicated in OXA resistance using classical LM. These results validate use of X-QTL methods for an important human pathogen, and will greatly simplify genetic analysis of traits such as virulence and host specificity.

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ACCURATE EXOME SEQUENCING IN A MICROSCOPIC INVERTEBRATE

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Small parasites and larval stages pose a problem for population genomic analyses because limited amounts of DNA template are available, while the large size of many parasite genomes makes sequencing complete genomes prohibitively expensive. For example, schistosome adults live in human blood vessels and only microscopic larval stages (miracidia) are available for molecular analysis. Furthermore, sequencing of the complete 363Mb genome is impractical for population samples. We evaluate the accuracy of exome sequencing of single miracidia following whole genome amplification and exome capture using a custom Agilent SureSelect array designed to capture 92% of the 15.3Mb exome. Because the miracidial exomes sequenced were obtained from a cross between a male and female worm of known sequence, we were able to predict the SNP alleles present in the progeny and quantify the genotyping error rate. We were also able to evaluate bias in capture of alleles recovered from genome regions showing high levels of polymorphism compared with the reference genome from which the capture array was designed. In initial work we robustly scored 18,993 SNPs, while judicious multiplexing of samples prior to genome capture reduced costs while maintaining accuracy. We conclude that scoring of genome wide exomic SNPs and indels from exome sequence is feasible, economical and extremely accurate and has many advantages over other methods for generating reduced representation libraries such as RAD-seq. This approach will allow research on schistosomes (and other parasite species) to progress from population genetics using small numbers of markers, to population genomics utilizing genome-wide marker information.

UNDERSTANDING THE GENETIC BASIS OF DRUG RESISTANCE IN TRICHOMONAS VAGINALIS

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In light of antimicrobial role in shaping genetic variation, understanding the regulation and control of drug resistance is imperative. Trichomonas vaginalis is the most widespread non-viral sexually transmitted pathogen in the world: it is commonly treated with the 5-nitroimidazole group of drug with pronounced resistance prevalence of up to 9.5%. The causes underlying resistance in T. vaginalis remain unknown. Our earlier studies of population structure in 46 clinical isolates using 21 microsatellite loci showed the drug resistance phenotype associated with genetic Type. This suggests a genetic component to 5-nitroimidazole resistance, although which genes are involved and whether recombination plays a role in acquiring drug resistance, remain to be determined. We started our studies by phenotyping a further 141 global isolates of T. vaginalis for metronidazole resistance using a minimum lethal concentration assay. Resistance was observed as follows: New York City: 22%; Mexico: 9%; Papua New Guinea: 8%; Africa: 4%; no resistant isolates were identified in 10 and 12 isolates from Australia and Italy, respectively. To obtain a detailed view of genome-wide variation in these total 187 isolates, we used double digest Restriction-Site Associated DNA (ddRAD) sequencing, which allows for partial genome sequencing and thus enables the highly repetitive regions of the T.vaginalis genome to be avoided. Using EcoRI (found to cut 5 times more frequently in unique regions of the genome) together with NIaIII enzymes for ddRAD library construction, we generate sequence data for 48 metronidazole resistant and sensitive T. vaginalis isolates using the Illumina HiSeq. The reference genome assembly and annotation served as a control. On average, 50% of highquality reads mapped back to the reference assembly. A total of ~6,900 unique gene regions from ~57,000 were sequenced to 20X or more coverage. Variant discovery to uncover the genetic basis of drug resistance are in progress.

HORMESIS AS A RESISTANCE ENHANCER IN STENOTROPHOMONAS MALTOPHILIA: WHICH CONSEQUENCES FOR CLINICAL THERAPY?

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Opportunistic pathogens are exposed to antimicrobial subinhibitory concentrations in nature and human body compartments during antibiotherapy. According to hormesis concept, although high therapeutic antimicrobial levels inhibit pathogens growth, subinhibitory concentrations could have a stimulatory effect in growth. Hence, this work aimed to probe bacterial growth stimulation by antimicrobials and assess if such stimulatory concentrations could enhance resistance in intrinsically resistant opportunistic pathogens, environmentally sourced, as *Stenotrophomonas maltophilia*. Using *S*. maltophilia D457 strain and a derivative mini-Tn5-mutant library, minimal stimulatory concentrations (MSCs) and minimal inhibitory concentrations (MICs) were established and used in evolution experiments performed until 157 generations. In accordance with hormesis, bacterial growth was stimulated under subinhibitory concentrations, at MSCs 16, 160 and 64 fold lower than MICs of norfloxacin, chloramphenicol and carbenicillin, respectively. Continuous exposure to MSCs during 157 generations lead to MICs 4 fold higher for norfloxacin, and 3 fold higher for chloramphenicol and carbenicillin, in comparison with evolution in antibiotic absence, and MICs 8, 6 and 4 fold higher, in comparison with initial generation. Exposures to MSCs during 9 or 157 generations lead to similar resistance levels. Thus, our results suggest that stimulatory concentrations, present in nature or human body compartments, might lead to the swift enrichment or emergence of resistant mutants, contributing to therapy failure as well as to the rapidity of epidemiological spread of S. maltophilia resistance determinants in clinical environments. This work highlights the importance of hormesis to be included in pharmacokinetic-pharmacodynamic modelling of existing and newer antimicrobials, to predict the impact on the rapidity and potential development of antimicrobial resistance in such relevant opportunistic pathogen.

D20SY22PS1035

MODELING RESISTANCE TO CHEMOTHERAPEUTANTS IN SALMON ECTOPARASITES: THE INFLUENCE OF TREATMENT REGIMENS AND TEMPERATURE

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Sea lice (Lepeophtheirus salmonis) are ectoparasites of wild and farmed salmonids. On salmon farms they are often controlled using integrated pest management (IPM) programs that include the application of chemotherapeutants. Resistance to chemotherapeutants typically appears on farms after less than a decade of chemical use. Infestations are influenced by environmental factors including temperature. In warmer temperatures, sea lice generation times are shorter and the net reproductive rate is greater. While it is likely that life history changes of this type also increase the rate at which pesticide resistance evolves, this has never been tested. To understand how temperature and IPM programs influence resistance evolution, we constructed individual-based models of a sea lice population that experiences periodic selection by a pesticide. There is support for several different mechanisms of chemical resistance in sea lice and we explore how outcomes differ when resistance is caused by a single gene with large effect in comparison to many genes with small effects. We find that the rate at which resistance evolves is sensitive to temperature, treatment regimens, and the life history stage(s) being targeted for control.

D20SY22PS1122

MANIPULATION OF THE WITHIN-HOST ENVIRONMENT PREVENTS THE EMERGENCE OF RESISTANT PARASITES

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Aggressive, high-dose drug treatment has been advocated as a resistance management strategy because it dramatically reduces the population size of parasites and thus the probability that a *de novo* resistance mutation will emerge. However, in mixed genotype infections aggressive drug treatment confers a great selective advantage to any pre-existing drug-resistant parasites and enables their growth by releasing them from competition with drug-susceptible parasites. We attempted to resolve this tension by mimicking the suppressive presence of susceptible competitors by manipulating the availability of a resource necessary to parasite growth. Mice were inoculated with both susceptible and resistant strains of the rodent malaria Plasmodium chabaudi and either provided with a nutrient or deprived of it. Following aggressive antimalarial treatment, resistant parasites in nutrient-supplemented mice grew to densities of ~1 million per microliter, causing a concurrent bout of anaemia. In contrast, no resistant parasites were observed in nutrient-deprived mice, which remained healthy. We concluded that this effect was mediated by the combination of competition and nutrient deprivation since resistant parasites grew well in single infections, irrespective of nutrient treatment. We propose that environment-manipulating compounds, which may go undetected in screens for lethal drugs, might present a novel class of drugs and that this approach is a novel, potentially 'evolution-proof', means of slowing the emergence of resistant pathogens.

D20SY22PS1168

PHYLOGENETICS OF HIV DRUG RESISTANCE: CAN RESISTANT STRAINS COMPETE?

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The emergence of drug resistant HIV is a major challenge facing efforts to prevent AIDS and new infections through widespread drug treatment. Much work has focused on the patterns of treatment failure and evolution of drug resistance in infected individuals, but this does not directly correspond to drug resistance evolution in a population. HIV sequences from infected individuals can be used to identify drug resistance mutations, and to reconstruct phylogenetic relationships between viruses, allowing inference of the transmission chain (i.e. the phylogeny). We use HIV *pol* data from the Swiss HIV Cohort Study to estimate the relative rates of emergence and transmission of drug resistant virus. The inferred phylogeny and drug-resistant status of each individual is fitted to a two-strain birth-death model which quantifies the fitness cost in transmission of drug-resistant strains. Initially the methods have been tested by simulating transmission chains with drug resistance evolution, and then attempting to re-estimate these values. Substantial correlation between the parameters present challenges to their estimation, and indicate that the methods require further development. However, initial results from the data suggest that the basic reproductive number of resistant virus in this cohort is substantially lower than that of the drug sensitive virus. The evolution of virus from sensitive to resistant is a much more common event than reversion to the sensitive type. Drug resistant virus appears to spread too slowly to produce a self-sustaining epidemic, and is instead continually seeded from an ongoing drug sensitive epidemic. However, further refinement of the methods are needed to justify this conclusion. A future key step is to investigate the spread of resistant viruses in high prevalence areas, where drug treatment is increasing, but without the frequency of testing and regime flexibility offered in resource-rich areas.

D20SY22PS1261

MECHANISMS OF BALANCING SELECTION FOR ANTIBIOTIC RESISTANCE IN STREPTOCOCCUS PNEUMONIAE

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Selection for antibiotic resistance shapes the evolution of many human pathogens, and understanding how resistant phenotypes arise and spread remains a challenge for evolutionary models. *Streptococcus pneumoniae*, or pneumococcus, is a common asymptomatic colonizer of the human nasopharynx. Over the past few decades, strains of pneumococcus have emerged that are resistant to several major classes of antibiotics. Curiously, the resistant phenotypes appear to be stably coexisting with sensitive strains. Previous models have suggested that such coexistence is unlikely in the absence of implausibly high levels of population structure or intra-strain competition. Here, we use a computational model that explicitly incorporates known mechanisms of immune-mediated competition to evaluate these and other hypotheses. We find an expanded set of scenarios that support balancing selection, several of which may shape the coexistence of resistant and sensitive strains in other species.

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MACROEVOLUTIONARY DYNAMICS OF INTRINSIC ANTIBIOTIC RESISTANCE IN *PSEUDOMONAS*

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The anthropogenic use of antibiotics has driven the rapid evolution of high levels of resistance, but the extent to which antibiotics imposed selection for resistance in the pre-human era remains largely unknown. Using a novel comparative approach to study resistance, we show that selection for low levels of antibiotic resistance ("intrinsic resistance") is ancient in bacteria from the genus Pseudomonas. At a phenotypic level, we show that divergence in intrinsic resistance between species does not correlate to evolutionary distance, suggesting that the evolution of resistance is driven by selection, and not neutral drift. Consistent with this argument, we find evidence of a genome-wide trade-off between intrinsic resistance and competitive ability, implying that selection is required to maintain resistance. Recent work has shown that hundreds of genes with diverse functional roles contribute to intrinsic resistance, but we show that approximately 80% of the phenotypic variation in resistance between species is determined by variation in a small number of genes with known roles in protection against antibiotics, most of which are antibiotic efflux pumps. Importantly, these are the same genes that provide the dominant mechanism for the evolution of resistance to clinical doses of antibiotics in *Pseudomonas*. In summary, our study shows that antibiotics represent an ancient evolutionary pressure, suggesting that historical patterns of evolution of resistance are likely to play a key role in determining the potential for evolving resistance to clinical doses of antibiotics.

D20SY22PS1332

INVASION OF FUNGICIDE RESISTANCE: IS SPATIAL SCALE IMPORTANT?

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Fungicide resistance is an urgent problem in agriculture. It makes the chemical control of fungal diseases of plants ineffective and leads to sever crop losses. In order to manage fungicide resistance, a thorough understanding of invasion, persistance and spread of fungicide-resistant pathogen strains is required. At the same time, the dynamics of fungal diseases of plants is strongly influenced by spatial scales of the host-pathogen interaction: spatial extension of the host population and the dispersal range of the pathogen. Here, we explore theoretically the effect of these characteristic spatial scales on the invasion of fungicide resistant pathogen strains using a population dynamics modeling framework.

D20SY22PS1456

HEAVY METAL DRIVEN CO-SELECTION OF ANTIBIOTIC RESISTANCES IN AQUATIC ENVIRONMENTS AND AN EVOLUTION BASED CONCEPT OF RISK ASSESSMENT

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Recently, it has been realized that an important part of the dissemination of antibiotics and the evolution of antibiotic resistant microbes depends on aquatic environments. In aquatic environments, bacteria from different origins (human, animal, environmental) are able to mix in the presence of antibiotics or other pollutants such as heavy metals. Thus, the objective of our study was to identify which heavy metals potentially induce co-selection for antibiotic resistances in aquatic environments. Therefore, the available data on heavy metal driven co-selection in aquatic environments was reviewed and an evaluation criterion named minimum co-selective concentration (MCC) was developed. The MCC describes the lowest heavy metal concentration which was detected in correlation with increased bacterial antibiotic resistances in the environment. Additionally, a case study at the Western Bug River (Ukraine) investigating heavy metal concentrations of river water and sediment as well as antibiotic resistance of isolated Pseudomonas and Aeromonas strains was conducted. The results of both, the literature and the case study, indicate that metals such as mercury (Hg), cadmium (Cd), copper (Cu), and zinc (Zn) reach aquatic environments and accumulate to critical concentrations that trigger coselection for antibiotic resistance. Hence, future risk assessments for antibiotic resistance evolution should move from a clinical perspective towards an environmentally based assessment, considering sublethal concentrations of selective pollutants such as heavy metals. The here presented approach also considers evolutionary processes such as selection, co-selection and mutation. This approach towards a novel concept for risk assessment will be pivotal for detecting antibiotic resistance evolution in the environment.

Symposium

23. Genomic Islands: Their Role in Adaptation and Speciation

20 and 21 August



Program

Tuesday 20 August Session(s): 1, 2, 3, 4 Wednesday 21 August Session(s): 5, 6

Organisers: Rui Faria, Sarah Helyar, Chris Jiggins, Gary R. Carvalho,

Arcadi Navarro and Carole Smadja

Invited speakers: Jeffrey Feder, Nicholas Barton and Patrick Nosil

Description:

Genome-wide data offers a unique opportunity to gain an insight into the genomic architecture involved in the adaptive divergence found in heterogeneous environments which can lead to local adaptation, reproductive isolation and ultimately speciation. By considering a range of species and approaches, this symposium will explore the interactions of diversifying selection and homogenizing gene flow to identify trends in genomic evolution which accompany adaptation and ecological speciation. The symposium will focus on both theory and empirical data to assess the importance (or otherwise) of genomic islands of divergence.

23. Genomic Islands: Their Role in Adaptation and Speciation

D20SY23IT10:30R3

RAPID GENOMIC DIVERGENCE ACROSS THE SPECIATION CONTINUUM

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A new genomic theory for population divergence is presented predicting rapid speciation-with-geneflow when a critical threshold of divergently selected loci is reached. The theory is explored as a possible explanation for recent findings from several model speciation systems implying that extensive genetic differentiation may characterize even early stages of ecological divergence. In particular, patterns of genomic divergence among Rhagoletis fruit flies - a model for ecological speciation-withgene flow via sympatric host plant shifting - are examined as a test for the theory. If true, the theory provides a means to account for how gradual adaptive change, as epitomized by Darwin, can be reconciled with observations of sudden bursts of speciation, as occur during adaptive radiations, without the need to invoke macroevolutionary processes.

23. Genomic Islands: Their Role in Adaptation and Speciation

D20SY23IT14:00R3

WHAT CAN WE LEARN BY SAMPLING GENOMES ACROSS CLINES?

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Allele frequency clines reflect selection, whilst linkage disequilibria reflect patterns of gene flow and introgression. However, such patterns are noisy, because of sampling drift, patchy habitat, and the history of the cline; this makes it difficult to detect selection and gene flow by scanning across the genome. I review the inferences that can be made from genomic patterns of Fst, cline width, cline concordance, and introgression.

D21SY23IT10:30R3

EXPERIMENTAL INSIGHTS INTO THE PROCESSES DRIVING GENOME EVOLUTION

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Understanding the processes driving patterns of genome variation is a major goal in biology. Although observational genome scans have usefully quantified heterogeneity in levels of genetic differentiation across the genome, it can be problematic to infer evolutionary process and causation from such genome scans alone because a particular pattern of genomic divergence could arise via various combinations of selection, recombination, drift, gene flow, mutation, and demographic history. In this regard, experiments have the potential to isolate the contributions of specific evolutionary processes to patterns of genome variation. In this talk, I describe the processes driving and constraining genome evolution in experimental populations of stick insects that were transplanted to novel host-plant and climatic environments in the wild. I contrast the experimental results with genome variation among long-established natural populations and discuss the collective empirical results in light of theory. The talk will illustrate how combined experimental and genomic approaches can move the field of genomics towards becoming a more predictive science.

D20SY23RT11:18R3

HOW DOBZHANSKY-MULLER INCOMPATIBILITIES ACCUMULATE IN PRESENCE OF GENE FLOW?

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Interest in speciation research has experienced a recent shift from the classical problem of "When does it happen?" to more process-oriented questions: "How does it happen?" This is of relevance, in particular, for parapatric speciation, where the build-up of pre- or postzygotic barriers to gene-flow is a gradual process. The standard mechanism for the evolution of postzygotic isolation is the accumulation of Dobzhansky-Muller incompatibilities (DMI). While this process is reasonably well understood for allopatric speciation, one can ask how it unfolds in the face of gene flow. In a recent paper, Bank et al. (2012) have studied the very first step of this process and described the conditions for a first two-locus DMI to appear and be maintained. Here, we extend this model to study more than one DMI. In particular, we are interested in the influence of the presence of a first DMI on the fate of a second one and in predictions about the genetic architecture of the growing barrier. Using a combination of analytical and numerical methods, we analyze a migration-selection model with unidirectional gene flow from a continent to an island. As expected, we generally find that the presence of a first DMI makes it easier for further DMI's to be stably maintained – once it is established. However, the picture is much more complex for the establishment process of the second DMI itself. Depending on linkage patterns and the strength of the incompatibilities, the presence of the first DMI may either facilitate the origination of a second one or impede it. We interpret our results in the light of recent ideas of "islands of speciation" or "genome hitch-hiking."

D20SY23RT11:42R3

THE GENOMIC LANDSCAPE OF SPECIATION IN THE REPEATED ADAPTIVE RADIATIONS OF NICARAGUAN CRATER LAKE CICHLID FISHES

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Understanding the factors that drive and constrain populations to build up reproductive barriers and diverge is a key objective in evolutionary biology. Based on the framework of ecological speciation, next-generation-sequencing tools allow researchers today to investigate the dynamics of divergent selection and gene flow at a genomic level. Although this endeavor has been very fruitful for our understanding of speciation, no generalities have emerged so far and more empirical data from different taxonomic groups and ecological settings are needed. One of the main questions, for example, concerns the importance of the temporal succession of restricted genomic islands of divergence (divergence hitchhiking) versus a genome-wide progression of divergence (genome hitchhiking). Having colonized several isolated crater lakes from the same source population, Midas cichlid fishes (Amphilophus cf. citrinellus) in Nicaragua have diverged into convergent ecotypes recently, repeatedly, and in sympatry; features that make this an interesting setting for studying the genomics of ecological speciation. Furthermore, the partly repeated and probably still ongoing radiations are at different stages of divergence, which makes this system one of the rare cases to investigate the temporal succession of this process. Here we investigate genomic divergence in the entire Midas cichlid species complex using double-digest restriction-site-associated-DNA sequencing (ddRAD-seq). With 700 individuals our data set covers the whole currently known diversity of the species complex and allows for a multitude of meaningful population genomic comparisons. By mapping the thousands of generated RAD loci on a preliminary Midas cichlid genome assembly, we are able to address questions concerning the genomic landscape of repeated but differently progressed speciation events and provide candidate regions that might harbor the genetic basis of adaptation and speciation in the Midas species cichlid complex.

D20SY23RT14:48R3

ISLANDS OF GENOMIC DIVERGENCE ACROSS THE HOODED AND CARRION CROW HYBRID ZONE: LINKING GENES AND 'MAGIC TRAITS'

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A comprehensive understanding of the speciation process at a genomic level requires investigation at different stages in the evolutionary continuum from onset to completion and hybrid zones provide such opportunities. The European crow hybrid zone between the all black carrion crow (*Corvus* [*c*.] *corone*) and grey coated hooded crow (*C*. *[c.] cornix*) is a classic example of early stage speciation characterized by a lack of neutral genetic divergence, limited evidence of postzygotic reproductive isolation, and the rapid divergence of distinct plumage characteristics that mediate strong assortative mating. Here we took a population genomic approach to characterize genomic divergence between color morphs to identify genes with major effect on speciation and potential "magic traits" involved in the earliest stages of divergence. Initial analyses of 66 re-sequenced genomes reveal a heterogeneous landscape of genome divergence with 4 narrow regions of the genome that show significant differentiation between the grey hooded crow populations from Sweden and Poland with respect to black carrion crow populations from Germany and Spain. These regions are potentially associated with structural variants (i.e. a pericentric inversion on chromosome 18), as well as differentially expressed candidate genes within the melanogenic pathway. However, fixed differences detected in these genomic regions in contrast to no divergence of the coding sequence of MC1R or intron regions linked to primary candidate genes points to potential cis-regulatory elements driving the initial divergence of expression profiles and plumage characteristics. Comparison of outlier regions with both a white coated and all black outgroup species (*C. torquatus* & *C. brachyrhynchos*, respectively) will determine whether the divergence is the result of standing genetic variation or derived mutations.

D20SY23RT15:12R3

ARE ROBERTSONIAN FUSIONS BARRIERS TO INTROGRESSION IN A HOUSE MOUSE HYBRID ZONE?

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Genomic regions of low recombination are expected to contribute to the accumulation of incompatibilities allowing speciation with gene flow to occur. While chromosomal inversions are well documented in such processes, the role of Robertsonian (Rb) fusions (that join two acrocentric chromosomes by their centromere to form a metacentric one) is less clear. The house mouse is an emblematic model of Rb variation since one subspecies, M. m. domesticus, displays >80 Rb races. Previous cytogenetic studies demonstrated a decrease in recombination rates along the proximal 50% of Rb chromosomes in both homozygotes and heterozygotes. This study explores the role of Rb fusions in reducing gene flow by investigating their effect on genome admixture between two subspecies of the house mouse. In Denmark where the two taxa form a tension zone, M. m. domesticus carries 3 Rb fusions while M. m. musculus has an all-acrocentric karyotype. Genome admixture was assessed using 127 SNPs diagnostic of the two subspecies and covering the proximal 25% of 3 chromosomal arms involved in Rb fusions and 2 acrocentrics. We compared patterns of genomic admixture in the proximal regions to those at 86 diagnostic loci randomly distributed in the genome by fitting a genomic cline model with 3 parameters (centre and slope in each subspecific context). In the domesticus genomic context, Rb clines were much steeper than that of the average reference loci. Their centre was displaced in favour of musculus, however, due to introgression of large chromosomal blocks, and both of these patterns were stronger for the most proximal loci. This is compatible with the predicted effects of reduced recombination in Rb fusions coupled with incompatibilities in these genomic regions. In addition, clines for acrocentrics were smoother than average in the musculus genomic context, suggesting that other undescribed effects are acting, such as a possible advantage of domesticus acrocentric centromeres.

D20SY23RT15:45R3

SELECTION ON FLOWER COLOUR IN A HYBRID ZONE REVEALS INTERWOVEN PATTERNS OF GENOMIC DIVERGENCE AT A LINKED CLUSTER OF LOCI

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To understand how selection establishes reproductive barriers between populations, we need to identify the loci regulating the selected traits. Knowing that, how are sets of co-adapted alleles maintained when gene-flow occurs between distinct populations? One view is that genetic linkage maintains fit haplotypes, resulting in a genomic signature of selection as an "island of divergence". To address these issues we study a hybrid zone between two Antirrhinum species with different flower colours. Various phenotypes result from this hybridization, but are restricted to a narrow region (~1km) and do not spread to the parental populations. A gene regulating magenta flower pigmentation, ROS, was found to have a steep cline correlated with a flower colour cline, suggesting this trait is under selection. A second tightly-linked locus also regulating this trait, EL, has now been mapped. A genomic comparison of diversity between the Antirrhinum species shows a signature of selection in the region containing both loci (ca. 180kb~0.9cM). Surprisingly, this "island of divergence" includes non-shared and shared polymorphisms, questioning how the parental ROS-EL haplotypes are maintained in face of gene-flow. By genotyping plants across the hybrid zone we show that even though LD between them is high (r~0.9), recombinants do occur (~12% in the zone). This suggests that gene-flow between the two populations was enough to break fixed differences within the "island", resulting in a rugged divergence profile that includes fixed polymorphisms (linked to the functional loci) interspersed with shared ones. To our knowledge this is one of a few cases where a cluster of loci under selection has been finely mapped and its behaviour studied at a population-level. This analysis empirically demonstrates the significant impact gene-flow has in breaking associations of clusters of putatively co-adapted alleles, providing key insights to our interpretation of genomic signatures of selection.

D20SY23RT16:09R3

PATTERNS OF GENOMIC DIVERGENCE ACROSS PARALLEL HYBRID ZONES OF MIMETIC HELICONIUS BUTTERFLIES

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The butterflies *Heliconius erato* and *Heliconius melpomene* are well known for their amazing mimetic similarities in colour pattern. These colour patterns vary across their geographic distribution with parallel hybrid zones in both species found between adjacent colour pattern races. Here we use RAD sequencing to investigate patterns of genome-wide divergence across two such hybrid zones of both species, in Peru and Ecuador. These data are successfully used to map loci controlling colour pattern differences between races and confirm previous findings that largely the same loci are responsible for colour pattern differences in the two mimetic species. In addition, these loci are among a handful of divergence outliers identified in the genomes, supporting the assertion that strong selection acting on colour pattern is one of the primary forces maintaining these hybrid zones. In spite of the striking parallels at the genomic level between species we do also find some previously unidentified differences. We also compare different techniques for analysis of this type of data, which we envisage being ever more widely used for the analysis of adaptive divergence in wild populations.

D20SY23RT16:33R3

GENOMIC TESTS OF WHETHER CHROMOSOMAL REARRANGEMENTS FACILITATED LOCAL ADAPTATION IN ANOPHELES GAMBIAE BASED ON COALESCENT EXPECTATIONS

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As the primary malaria vector that is widely distributed in sub-Sahara, Anopheles gambiae s. str. utilizes many different microhabitats where human reside. Its fast adaptation is partly attributed to polymorphic inversions within or between populations in the species. Theory predicts that large paracentric inversions, which do not directly affect individual fitness, can facilitate local adaptation by suppressing recombination among co-adapted genes captured by inversions. Clines and associations between inversions and specific environmental factors are suggestive of causal relationships, but do not demonstrate that the inversions facilitated the local adaptation. Here, by comparing the time to coalescence of genomic regions within inversions and among collinear regions, we can estimate the age of inversions, selective strength acting on inversions in local populations, and how selection varies depending on the connectivity among populations. Genomic regions inside inversions where coadapted genes can be identified as well. In this study, wild mosquito samples were collected from five populations in transitional ecozones between forest and savanna in Cameroon. Karyotypes for two major inversions, 2La and 2Rb, were determined molecularly and genome-wide SNPs were identified for each individual from barcoded Illumina sequencing. Genetic data, together with spatial information on the sampled individuals, are then analyzed with a coalescent modeling framework to test whether inversions contributed to genomic divergence among ecologically dissimilar populations under gene flow. The approach provides new insights into genomic profiles when population divergence (or speciation) is promoted by structural genomic variants.

D20SY23RT16:57R3

MOSAIC GENOMES IN TIME AND SPACE REVEALED BY LARGE-SCALE TARGETED SEQUENCING IN MYTILUS MUSSELS

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Marine species with high rates of dispersal have often had complex histories of fragmentation and colonization. These can subtly modify the genomic barriers to gene flow between hybridizing populations, leading to mosaic patterns of genetic divergence. The *Mytilus* complex of marine mussels exemplifies these processes, as it includes populations with a wide range of divergences, and has a world-wide distribution, which has led to multiple independent contacts between the same species pairs in different ecological contexts. To understand patterns of genomic divergence in *Mytilus*, we conducted a large-scale targeted genomic capture experiment, investigating fine-scale variation in genetic diversity. Our target genomic regions included islands of divergence and valleys of introgression identified from Fst scans and BAC sequencing, as well as randomly chosen controls. Each region was sequenced in twelve genetic backgrounds from three *Mytilus* species, distributed throughout the northern hemisphere. By studying variation in gene genealogies, our results illustrate how genome mosaicism varies in time and space. We also compare nuclear coding genes of known function (including immunity, cyto-nuclear interactions and nucleoporins) to randomly chosen control genes.

D20SY23RT17:45R3

CROSSING THE SPECIES BARRIER: GENOMIC ISLANDS OF INTROGRESSION BETWEEN TWO EXTREMELY DIVERGENT CIONA INTESTINALIS SPECIES

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Inferring a realistic demographic model from genetic data is an important challenge to gain insights into the historical events during the speciation process and to detect the molecular signature of selection along genomes. Recent advances in divergence population genetics have reported that speciation in face of gene flow occurred more frequently than theoretically expected, but the approaches used rarely account for genome wide heterogeneity (GWH) in introgression rates. However, GWH is expected as a consequence of variation in effects of natural selection on migrant alleles. We investigated the impact of GWH on the inference of divergence with gene flow between two cryptic species of the marine model Ciona intestinalis. These morphologically indistinguishable entities are highly diverged molecular-wise, but evidence of hybridisation has been reported in both laboratory and field studies. We examined polymorphism and divergence patterns across 852 genes scattered throughout the C. intestinalis genome. We compared various speciation models and statistically tested for GWH under the ABC framework. Our results demonstrate the presence of significant extents of gene flow resulting from a recent secondary contact between the two gene pools, after more than 3My of divergence in isolation. The inferred rates of introgression are relatively low, highly variable across loci and mostly unidirectional, which is consistent with the idea that numerous genetic incompatibilities have accumulated over time throughout the genomes of these highly-diverged species and that introgression could be adaptive. A genomic map of the level of gene flow identified two islands of introgression, i.e. large genome regions of unidirectional introgression. This study clarifies the history and degree of isolation of two cryptic and partially sympatric model species, and provides a methodological framework for the study of GWH in introgression rates at various stages of the speciation process.

D20SY23RT18:09R3

IDENTIFYING LOCI UNDER SELECTION AGAINST GENE FLOW IN ISOLATION WITH MIGRATION MODELS

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When divergence occurs in the presence of gene flow there can arise an interesting dynamic in which selection against gene flow, at sites associated with population specific adaptations or genetic incompatibilities, can cause net gene flow to vary across the genome. Loci linked to sites under selection may experience reduced gene flow and may experience genetic bottlenecks by the action of nearby selective sweeps. Data from histories such as these may be poorly fit by conventional neutral model approaches to demographic inference which treat all loci as equally subject to forces of genetic drift and gene flow. To allow for demographic inference in the face of such histories, as well as the identification of loci affected by selection, we developed an isolation with migration model that explicitly provides for variation among genomic regions in migration rates and/or rates of genetic drift. The method allows for loci to fall into any of multiple groups, each characterized by a different set of parameters, thus relaxing the assumption that all loci share the same demography. By grouping loci, the method can be applied to data with multiple loci and still have tractable dimensionality and statistical power. We studied the performance of the method using simulated data, and we applied the method to study the divergence of two subspecies of European rabbits (*Oryctolagus cuniculus*).

D20SY23RT18:33R3

THE LANDSCAPE(S) OF GENOME DIVERGENCE IN *FICEDULA* FLYCATCHERS

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Unravelling the landscapes of genome divergence between individuals, populations, and species is key to understand the genomic architecture of biological diversification, and the forces driving its evolution. In the present study we make use of the recently available flycatcher genome (Ellegren et al. 2012, Nature), to study the genomic divergence between the naturally hybridizing but reproductively isolated collared flycatcher (Ficedula albicollis) and pied flycatcher (F. hypoleuca). Population genomic analysis of each 10 genomes of both species reveal a highly heterogeneous landscape of genome divergence with about 50 'divergence islands' that show ~50-fold higher differentiation than the genomic background. The non-randomly distributed divergence islands are characterized by reduced nucleotide diversity, skewed allele-frequency spectra, elevated linkage disequilibrium and reduced proportions of shared polymorphisms in both species, indicating parallel episodes of selection. The high incidence of divergence islands with genomic regions resistant to sequence assembly, potentially including centromeres and telomeres, indicate that complex repeat structures may drive species divergence. In order to identify the molecular and evolutionary forces driving the evolution of divergence islands in Ficedula flycatchers, these insights are complemented with recent results from population genomic analyses of an additional 200 flycatcher genomes from multiple populations and

species.

D20SY23RT18:57R3

THE GENOMICS OF BIOLOGICAL CLOCKS: LOCAL ADAPTATIONS IN DIURNAL AND LUNAR EMERGENCE TIME OF THE INTERTIDAL MIDGE CLUNIO MARINUS (CHIRONOMIDAE, DIPTERA)

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The marine midge *Clunio marinus* (Chironomidae, Diptera) is found in the rocky intertidal zone of the European Atlantic coast. Reproduction is timed to the tides by means of diurnal and lunar rhythms. As tidal regimes differ along the coast, populations of *Clunio marinus* display a variety of local genetic adaptations in diurnal and lunar timing. These adaptations offer unique opportunities to study the evolutionary forces that shape local adaptation in a one-dimensional habitat, the coastline. Population genomic analysis revealed that the timing adaptations evolved within the last 20,000 years. Clunio marinus is restricted to rocky coasts. Populations from distinct rocky coasts, which are separated by long sandy stretches (e.g. Spain and the English Channel), are geographically isolated. This facilitated the evolution of timing adaptations. However, within stretches of continuous rocky coast there is little genetic differentiation between populations and timing adaptations are likely kept up by permanent local selection on the decisive loci. Crossing experiments, linkage mapping and QTL analysis revealed that the timing adaptations are controlled by only three major effect loci, the effective genes within these loci still being unknown. Some of the QTLs are associated with large chromosomal rearrangements and regions of suppressed recombination. After assembling the *Clunio marinus* genome, population samples of the strains used in QTL mapping were re-sequenced and screened for strainspecific SNPs, insertion-deletions, inversions and transposable element insertions. Genome wide, this identified >600,000 genetic polymorphisms, but less than 1,000 of these sites have strain-specific alleles. Less than 100 strain-specific variants are within the QTLs, pointing to the decisive genes within the genomic islands of adaptation. Currently, the resulting candidate genes are being tested for their functional role in the local timing adaptations.

D21SY23RT11:18R3

QUANTIFYING THE NEUTRAL EXPECTATIONS OF GENOMIC PATTERNS OF DIVERGENCE

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With decreasing costs of sequencing, we see a tremendous accumulation of genomic data from natural populations and non-model organisms in particular. However, a rigorous understanding of the processes that determine the evolution of genomic islands of divergence is still incomplete. Moreover, we lack well-defined neutral expectations of genome-wide differentiation, making it hard to distinguish neutral patterns from true signatures of selection. To help theory keep up with the rapid accumulation of data, we use coalescent simulation modeling of neutral divergence with gene flow to fully characterize genome-wide distributions of Fst. Mean, variance and skew of Fst across the genome depend strongly on migration and mutation rates. In addition, autocorrelation in divergence along the genome resulting from physical linkage can lead to neutral patterns that strongly resemble genomic islands. These findings challenge our interpretations of genomic variation in the context of ecological divergence. A more comprehensive baseline of neutral expectations will help determine which statistics describing genomic patterns have the most power to distinguish genomic islands of divergence resulting from selection as opposed to neutral processes.

D21SY23RT11:42R3

THE INFLUENCE OF THE GENETIC ARCHITECTURE OF A TRAIT ON LOCAL ADAPTATION IN A SUBDIVIDED POPULATION

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We investigate the consequences of spatially varying, stabilizing or directional selection on a quantitative trait in a subdivided population. The influence of the genetic architecture of the trait on local adaptation and on differentiation is explored by employing a deterministic two-locus, two-deme model. Our analysis demonstrates that the critical migration rate, below which local adaptation is possible, depends crucially on the genetic architecture of the trait. Under stabilizing selection, but with different optima in the demes, strong recombination may facilitate local adaptation by enhancing genetic variability. In contrast, with directional selection in opposite direction, local adaptation is maximized by a concentrated genetic architecture, i.e., by a major locus and a tightly linked minor one. Complementing the work of Yeaman and Whitlock (2011), who found that concentrated genetic architectures may evolve in subdivided populations under directional selection, we showed that such architectures considerably facilitate local adaptation and increased differentiation.

D21SY23RT14:00R3

LOCAL ADAPTATION, INVERSIONS AND NON-GENIC VARIATION IN TEOSINTE

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Populations of widely distributed plant species have to adapt to local environmental conditions. Locally adapted plant populations are often observed, but the genetic basis of adaptation is rarely known. We used high-density genotype data and a wide set of environmental variables to identify adaptive genetic variation in teosinte, a wild relative of maize. Overall, population structure in teosinte was strong and was affected by altitude, dispersal events and admixture between subspecies. Patterns of linkage disequilibrium revealed four megabase-scale inversions that were polymorphic in teosinte populations. Inversions segregated in clinal patterns, were associated with environmental variation and were extremely differentiated among subspecies and populations. In addition, we found that non-genic variation plays a role in local adaptation of teosinte. These results suggest that large structural rearrangements and variation outside coding regions can be important for adaptation, especially in large plant genomes in which the gene space constitutes a small fraction of the entire genome.

D21SY23RT14:24R3

POPULATION GENOMICS OF PARALLEL EVOLUTION IN THE TRINIDADIAN GUPPY (POECILIA RETICULATA)

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Often the clearest evidence we have of natural selection is when populations or species independently adapt to a similar environment in a similar way. Despite its importance, however, we often do not know the genetic architecture of such evolution. The guppy (Poecilia reticulata) offers an exciting opportunity to study parallel evolution. In northern Trinidad guppies below waterfalls co-exists with many predators, while guppies above waterfalls experience low predation. Guppies in separate rivers often evolve similarly to predation pressure independently, e.g. more colouration, larger size at maturity, and less shoaling in low predation localities. Furthermore, the guppy, offers another advantage because their natural history facilitates experimental studies in nature. Researchers have established experimental populations, by transplanting fish from high predation populations, to areas without guppies or predators. The evolution at the phenotypic level has been well documented in these populations, where guppies adapt to their new environment in four years or less. I am using a population genomics approach to identify selected alleles in high and low predation population pairs and experimentally established populations. I genotyped approximately 20 individuals each from 14 populations using a RAD-Seq approach. The reads were sequenced on an Illumina HiSeq 2000, 100 bp single end. A total of 967 million reads were generated. The reads were then filtered for sequence quality, sorted for unique barcode, and mapped to a preliminary draft of the guppy genome. A signature of selection is detected when SNPs are more diverged at specific loci then what would be expected by neutrality. Parallel evolution is detected when outliers occur in the same region in multiple pairwise comparisons. This study will be the first to examine the genetics underpinning local adaptation in this evolutionary model system.

D21SY23RT14:48R3

A NOVEL GENOMIC SIGNATURE OF ADAPTATION FROM STANDING VARIATION REVEALED IN THREESPINE STICKLEBACK

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Understanding how natural selection shapes the genome is becoming a major endeavor in evolutionary biology. Selection will often target pre-existing (standing) genetic variation. Nevertheless, theory has generally been developed for selection on novel variants. We here use extensive modeling tailored to adaptive divergence in threespine stickleback to characterize the genomic footprints of adaption from shared standing variation. The emerging predictions are then examined using genome-wide markers and targeted sequencing across many natural populations. Combined, our findings offer a novel explanation for heterogeneous genomic differentiation between diverging populations, challenge the widely held view that genomic regions of high and low population differentiation reliably point to loci under divergent and balancing selection, and allow us to propose a novel methodological framework for searching adaptation genes in natural populations.

D21SY23RT15:12R3

GENOMIC ISLANDS OF DIVERGENCE: A COMPARISON OF FIVE PARAPATRIC LAKE-RIVER POPULATION PAIRS OF THREE-SPINED STICKLEBACKS

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Sticklebacks have recently colonized various freshwater habitats and have since undergone recurrent phenotypic divergence. Here, we sample five geographically distinct lake-river population pairs of the three-spined stickleback for investigating patterns of population divergence at varying stages of ecological speciation. Using 60 whole genome sequences (15x coverage each) we identify genomic islands of divergence between parapatric lake-river ecotypes. This allows us to evaluate the number, size and distribution of genomic islands of divergence between several population pairs and to assess the extent of population differentiation across our samples. We identify different genes and genomic regions potentially associated with ecological differentiation in the five lake-river comparisons. This suggests that divergence patterns between lakes and rivers can involve multiple molecular pathways. Interestingly, we find some associations between genomic structural variations and islands of divergence. This is based on our extensive set of structural variations including deletions, copy-number variations, inversions, and translocations. Combined with estimates of the recombination rates across the genomes, we provide insights into the relevance of the genomic architecture and the role of genomic islands during adaptive divergence.

POSTERS

D20SY23PS0016

FST OUTLIER TESTS VS. ENVIRONMENT-ALLELE ASSOCIATIONS IN GENOME SCANS FOR LOCAL ADAPTATION: WHEN DO THEY TELL THE TRUTH AND WHAT ARE WE MISSING?

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Next-generation technology has made it possible to obtain large amounts of genomic data. But how do we find locally-adapted genes from this mountain of data? One method is to look for outliers in the distribution of FST (FST outlier tests, FDIST (Beaumont and Nichols 1996) and BAYESCAN (Foll and Gaggiotti 2008)). Another method is to look at the correlation between allele frequencies and the putatively selective environment (environment-allele associations or EAAs, such as SAM (Joost et al. 2007) and BAYENV (Coop et al. 2010)). Using a large-scale landscape genetics simulator, we compared these two approaches for four common demographic histories with the same mean FST: island model, isolation-by-distance, expansion from one refuge, and expansion from two refugia with secondary contact. The latter two demographies were non-equilibrium scenarios. We found a widely used EAA called SAM doesn't control for population structure and has an unacceptable rate of false positives. For the refugia scenarios, we found that FST outlier tests had more false positives, but BAYENV had few false positives. In contrast, for isolation-by-distance, we found that FST outlier tests resulted in few false positives, but BAYENV had many false positives. We explored the range of conditions over which FST Outlier Tests and EAAs could detect loci under selection, and we show that each method has different power depending on demographic history. We propose a decision process that can used to identify demographic histories that are likely to cause high error rates. We also suggest two approaches that can be used to more accurately account for demographic history in genome scans and EAAs.

D20SY23PS0020

THE ROLE OF SELECTION IN DRIVING LANDSCAPE GENOMIC STRUCTURE OF THE WATERFLEA DAPHNIA MAGNA

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Evolutionary changes are determined by a complex assortment of ecological, demographic and adaptive histories. Predicting how evolution will shape the genetic structures of populations has principally relied on investigations through space, in lieu of time, because long-term phenotypic and molecular data are scarce. Yet, dormant propagules in sediments, soils and permafrost are convenient natural archives of population-histories from which to trace adaptive trajectories along extended time periods. The water flea Daphnia is a renowned ecological model system that produces subfossil dormant egg banks. Capitalizing on this system we identify selective and demographic processes driving present-day population genetic structure by analyzing dormant eggs in space and time. We provide solid evidence of differential selection at the genome level under well-characterized environmental gradients in populations distributed at regional scale and identify candidate genes linked to three environmental stressors. We validate these spatial patterns in "time" on populations hatched from sediment cores with known history for the same three stressors and in experimental evolution trials. Building on these results, we study how the complex mosaic of biotic and abiotic variables naturally occurring in the landscape, in addition to the already studied gradients, contributes to driving neutral and adaptive genomic variation. We infer past and recent demographic processes by contrasting patterns of local and regional neutral genetic diversity at markers with different mutation rates. We observe that selection plays a major role in determining the population genomic structure of D. magna. Environmental selection directly impacts genetic variation at loci hitchhiking with genes under selection. Priority effects enhanced by local genetic adaptation (cf. monopolization) affect neutral genetic variation by reducing gene flow among populations and genetic diversity within populations.

GENETICS OF MIGRATION - INSIGHTS FROM A WILLOW WARBLER MIGRATORY DIVIDE

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Every autumn young passerines migrate to wintering areas without the guidance of experienced adults. This is made possible by the expression of a genetic program encoding information on when, in which direction and how far to migrate. Although it has been established that migratory behaviour has a strong genetic basis, virtually nothing is known about the underlying genes. Knowledge of specific migration genes would allow for a much deeper mechanistic understanding of migration. In addition, comparing the genes in different migratory organisms would shed light on the evolution of these behavioural adaptations. Our research focuses on two subspecies of the willow warbler Phylloscopus trochilus in Northern Europe. The subspecies are morphologically and genetically extremely similar, but migrate to different wintering areas in Africa. In order to detect genetic variation associated with their different migratory strategies, we used next-generation sequencing of expressed genes in migrating and breeding individuals of both subspecies. The sequence data suggests that a very small proportion of coding genes differ between the subspecies and that these genes are clustered in two chromosome regions. Genetic variation within these chromosome regions is currently being investigated in a hybrid zone between the subspecies. Because of the segregation of genetic variation in hybrids, the chromosome regions are expected to be more independently associated with the different migratory strategies. This will enable us to determine whether these chromosome regions are linked to migration.

ASSESSING PATTERNS OF HYBRIDIZATION BETWEEN NORTH ATLANTIC EELS USING GENOME-WIDE SINGLE NUCLEOTIDE POLYMORPHISMS GENERATED BY RAD-SEQUENCING

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Hybridization is an important issue in evolutionary biology and conservation. We investigate the dynamics of introgressive hybridization in North Atlantic eels, the European eel (Anguilla anguilla) and its sister species the American eel (Anguilla rostrata). The two North Atlantic eels occupy a broad range of habitats from Morocco to Iceland in the eastern Atlantic and from the Caribbean to Greenland in the western Atlantic, respectively. Despite such a wide non-overlapping geographic distribution, both species spawn in partial sympatry in the remote Sargasso Sea, in a migration of over 5,000 km for the European eel and 2,000 km for the American eel, which still represent to this day one of the enigmas of the animal kingdom. Spawning grounds of both species overlap in space and time, setting the stage for interbreeding. Previous studies have pointed to the existence of hybrids limited to one particular location, Iceland, although markers used did not allow clarification of their hybrid status. In our study, diagnostic species-specific single nucleotide polymorphisms (SNPs) are developed using a RADsequencing approach, which is a reduced representation genome sequencing method. After generating and aligning over 8000 sequence read per individual, a total of 3,348 diagnostic SNP (FST> 0.95) were identified. 96 of of those loci were selected to design a low-density array, at which we genotyped 159 eels from Iceland and 29 eels from Faroe Islands. The study was supplemented with the analysis of 94 larvae (49 European and 45 American eel) collected in the Sargasso Sea. Our study reveals high levels of hybridization in Iceland mostly consisting of F1 hybrids but backcrosses (A. anguilla x hybrid) are also observed. In comparison, no hybrids are observed in the closely-located Faroe Islands.

QUESTIONING THE STATUS OF THE TWO FAW (SPODOPTERA FRUGIPERDA, NOCTUIDAE) HOST RACES USING INTEGRATIVE APPROACHES

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The fall armyworm (FAW) Spodoptera frugiperda (Lepidoptera, Noctuidae) is a polyphagous pest responsible for serious damages on several crops in the Americas and Caribbean regions. The FAW is constituted of two strains that share the same distribution, one adapted to rice and one adapted to corn. Because adaptation to host plants is considered as a potential driver for ecological speciation, the FAW constitutes a classical model to study sympatric speciation. The two variants differ by their mating behavior, pheromone compositions, and present development variability according to the host plant. These evidences of reproductive isolation between the two strains suggest that the two variants may constitute two different species. However species status of S. frugiperda is still highly discussed because hybrids naturally occur in the wild and because of the controversial results concerning mating success between the two strains. Here, we present a combination of molecular and laboratory experiments performed in the aim of assessing the level of genetic differentiation between the two *S*. frugiperda strains. Species delimitation analyses based on coalescent approaches show that a wide sample of individuals from the two corn and rice strains, collected in natural populations, are segregated in two distinct putative species clusters. In addition, using microsatellites markers, and F2 inter- laboratory strains crosses of S. frugiperda, we found a high ratio (six out of 12) of markers departing from mendelian segregation, whereas only one microsatellite was distorted in intraspecific crosses. These two complementary approaches support a rather high level of genetic differentiation between the two strains. We will present preliminary analysis of genomic regions surrounding distorted markers (The FAW consortium, pers. comm.) which suggest putative involvement of candidate genes in inter-strain incompatibilities.

A GENOMIC APPROACH TO THE PLUMAGE COLOUR DIFFERENCES IN THE WILLOW GROUSE (LAGOPUS LAGOPUS)

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My work aims to combine the field of genetics with the application of population structure modeling as well as a more individual based genome modeling. The species of my study is the willow grouse (*Lagopus lagopus*), which has an interesting phenotypic diversity across its range. In Scandinavia, individuals adopt a white plumage during winter, while individuals in the northern UK forgo moulting and remain brown. Interestingly, snow free islands off the coast of Norway harbor the Scandinavian populations in fluctuating densities, which are seemingly maladapted seeing as their white plumage stands out. With the help of population modeling, one of the aims is to model the dynamics of movement to identify conditions required for local adaptation (i.e. what would it take for the island populations to stop their winter moulting). Furthermore, the genome of the willow grouse will be sequenced with the aim of finding potential genes responsible for this variation in moulting behaviour. In turn, models of the genomic data will be constructed to find out which parts of the genome are likely to respond to local selection pressures and random forces such as drift. Is it the case that local populations become divergent throughout the genome (as predicted by drift) or at key loci (as predicted by selection)? As such, the project aims to tackle these questions from an empirical as well as a theoretical perspective.

RAPID EVOLUTION IN THE WILD: UNRAVELLING THE GENETIC BASIS OF A WING MUTATION IN FIELD CRICKETS

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The field cricket *Teleogryllus oceanicus* is a textbook example of rapid evolution in the wild, but little is known about the genetics of this intriguing system. In Hawaii, a mutant variety of silent male crickets has recently arisen and spread in several populations, apparently in response to pressure from an acoustically-orienting parasitoid. The mutation, *flatwing*, first appeared on the island of Kauai in 2003, but it has since appeared on the neighbouring island of Oahu. It segregates as a sex-linked, sexlimited Mendelian trait in both populations, and it feminizes male wings by erasing the specialized sound-producing structures of a normal wing. The degree of feminization, however, varies between islands. Does this represent convergent evolution, or has gene flow introduced the mutation(s) into a different genomic background, resulting in different phenotypic manifestations? To pinpoint where the mutation or mutations reside within the genome, and to help answer this question, we applied Bulked Segregant Analysis (BSA) using RAD-seq in two different populations with distinct *flatwing* phenotypes and abundances: Kauai (>95% of males are *flatwing*, with heavily feminized wings), and Oahu (ca. 50% of males are *flatwing*, with less strongly feminized wings). We have identified the first group of candidate SNP markers associated with the *flatwing* phenotypes in each population. Using these, we tested different evolutionary hypotheses that could explain why there is a difference in mutant wing morphology between the two populations. The results provide insight into how convergent phenotypes arise and evolve in real-time, in the wild.

THE EFFECT OF LINKAGE ON SOJOURN TIMES AND PATTERNS OF GENOMIC DIVERGENCE

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The combination of spatially heterogeneous selection and gene flow can lead to clustering of adaptive mutations and concentrated genetic architectures. This might contribute to observed genome-wide variation in differentiation between populations or species, sometimes reflected in 'islands of divergence'. A mechanistic explanation is that tight linkage to previously established mutations reduces the effective migration rate experienced by novel mutations, which increases their potential of establishment. Deterministic and stochastic studies of invasion properties suggest that this benefit of linkage is constrained to a small genomic neighborhood. Other mechanisms such as so-called 'genomic hitch-hiking' or translocations might be needed to explain observed patterns. However, we argue that invasion properties alone yield an incomplete picture. With one-way migration and genetic drift, invasion of adaptive mutations is ephemeral. Therefore, it is essential to study the time a beneficial mutation spends in the population before loss. This sojourn time limits the potential for a further mutation to occur and profit from linkage. Using diffusion theory, we show that even under the strong assumption of quasi-linkage equilibrium, the presence of a previously established adaptive mutation may considerably elevate the sojourn time of a weakly beneficial mutation. Depending on the rate at which beneficial mutations occur, even loose linkage might therefore enhance their chance of invasion. In view of empirical studies, accurate estimates of both migration and recombination rates are necessary to judge if patterns of divergence and concentrated architectures can be explained by direct benefits of linkage to previously established polymorphisms.

POPULATION GENOMICS OF ADAPTIVE DIVERGENCE IN THREE-SPINE STICKLEBACK USING SEQUENCED RAD TAGS

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Local selective pressures exerted by environmental heterogeneity across the geographic range of species could act to maximize individual fitness within specific environments (local adaptation). Stickleback (Gasterosteus aculateus) inhabits a diversity of environments throughout the northern Hemisphere, ranging from relative stable marine environments and large lakes to small ponds, the latter of which are expected to represent stressful environments, particularly during hot summers. Here, we test the hypothesis that populations are locally adapted to their environment by analyzing footprints of selection along with possibly differences in morphological traits. We conduct a genome scan of nucleotide diversity and differentiation in natural Danish and Greenlandic populations of three-spine stickleback. We used Illumina-sequenced RAD tags to identify and type over several thousands of single nucleotide polymorphisms (SNPs) in each of 20 individuals per populations (marine versus freshwater populations). We specifically hypothesize that genes showing footprints of selection have functions that are of ecological importance in the specific environments. Hence, in shallow ponds experiencing warm summer temperatures, we would expect to see footprints of selection at genes related to thermal tolerance, such as heat-shock protein coding genes. We also expect to see footprints of selection at candidate genes for lateral plates in comparisons between freshwater and marine sticklebacks. In order to detect possible selection at the phenotypic level, we conduct morphological analyses of all sticklebacks subject to RAD sequencing. Finally, we estimate the divergence time between the freshwater populations and the marine ancestral populations, using neutral genetic variation to estimate the rates at which the adaptive responses have occurred.

GENETIC DIVERSITY AND HYBRIDISATION IN ANCIENT LIME TREES (TILIA)

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Hybridisation and introgression between species affect both speciation and adaptation, to varying degrees. Studies in, for example, oak and poplar have shown genomic regions that reveal high differentiation between species for regions with reproductive isolation genes and other regions with lower differentiation, containing adaptive genes. *Tilia cordata* (small leaved lime) and *T. platyphyllos* (large leaved lime) are endemic in Europe and they can hybridize (*T. x europea*). Although currently commercially not important, they play important roles in their ecosystems. A cooler climate and human impact has reduced sexual regeneration and the number of trees, especially in the north. However, genotypes can survive for many hundreds of years and individuals may therefore be ancient. Microsatellites markers confirm that the species are diploid with high diversity and a predominantly outbreeding mating system. It is unclear what the extent and direction of hybridisation and introgression are in the wild. There are some morphological differences, but in the absence of flowers it is often difficult to identify the species or the hybrid with certainty. Here we present preliminary studies investigating hybridisation in *Tilia*. The individuals clearly grouped by species, confirming their genetic differences. We found a significant number of hybrid individuals in some populations, confirming morphological classification in most cases. In subsequent studies we aim to use NGS technologies to understand better the genomics of speciation.

INFERRING THE HISTORY OF INVERSIONS USING SEQUENCED RESTRICTION-SITE ASSOCIATED DNA (RAD) DATA FROM NATURAL POPULATIONS OF MOSQUITOES

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Due to restricted recombination in heterokaryotypes, genomic regions close to inversions breakpoints constitute genomic islands where mutational differences can accumulate thereby facilitating adaptation in the face of gene flow. Anopheles baimaii is an abundant mosquito malaria vector in SE Asia, with cytological evidence for at least four segregating inversions throughout its distribution. To study the inversions, we analyzed pair-wise Linkage Disequilibrium (LD) data from 3197 sRAD loci from 147 wild-caught individuals using a novel approach based on social network analysis (SNA). Assuming some mutations have had time to accumulate on the inverted chromosome, loci close to its breakpoints are expected to show high LD when a population sample includes all karyotypes but not when karyotypes are analyzed separately. When only allowing relatively high LD values in SNAs, loci close to inversion breakpoints are expected to form tight clusters with high average numbers of edges (representing LD values above the cut off) per vertex (representing loci). Using this approach we extracted four LD clusters (LDCs) from the networks from which 33-15 loci each were analyzed further. For each LDC we used discriminate analysis of principal components to sort individuals into (two) putative homokaryotypes and a heterokaryotype. LD was non-significant when homokaryotypes were analysed separately and individual heterozygosity was over five times higher in the heterokaryotypes compared to homokaryotypes (for all LDCs), as expected if recombination is only restricted in heterokaryotypes. Geographical distributions of inferred karyotypes suggest selection as mechanism for maintaining polymorphism for two of the LDCs. Coalescent-based Bayesian ancestral reconstruction (BEAST) and isolation with migration (IMa2) analyses and suggest that all four of putative inversions have segregated in this species for a long time (100~200ky), potentially due to balancing selection.

COMPLETE MITOCHONDRIAL GENOMES OF THE BALTIC CLAM MACOMA BALTHICA (BIVALVIA: TELLINIDAE): VARIABLE GENE ARRANGEMENTS BETWEEN AND WITHIN LINEAGES AND GENETIC INCOMPATIBILITY

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Despite their large population sizes, high fecundity and dispersive potential, marine bivalves maintain adaptive polymorphism that help them cope with local environmental variations. The study of adaptive evolution can be facilitated by examining contact zones between close taxa. In these zones, maintained by endogenic genetic incompatibilities or exogenic barriers, hybrid genotypes persist despite a maladaptation state. The Baltic tellin *Macoma balthica* offers an opportunity to study the evolution of a hybrid zone within the context of contemporary climate change. A key species in intertidal trophic webs of northwest Europe, this bivalve seems to be affected by the warming of surface waters in the Bay of Biscay as its distribution range contracted 300 km poleward during the last thirty years. On European coasts, two lineages of *M*. *balthica* are present, and result from separate trans-arctic invasion events from the Pacific to the Atlantic Ocean, leading to strong admixture and hybridisation. How can these lineages maintain local adaptations? What are the genes implied? While screening for nuclear markers highly differentiated across populations, we found that the nuclear-encoded gamma subunit of the mitochondrial ATP synthase protein complex had high FST values, and may be undergoing adaptive selection. As proteins involved in the mitochondrial oxidative phosphorylation chain are prone to mitonuclear genetic incompatibilities and contribute to the maintenance of genetic barriers, we have set to sequence eight mitogenomes of *M. balthica*, describe gene content and genome organisation. Using long-range PCR, these mitogenomes were amplified and sequenced (MiSeq, Illumina). These results will help identify the mitochondrial genes under selective pressure by analysis of non-synonymous to synonymous mutation ratios, and test the hypothesis that the mitochondrial subunits of the ATP synthase complex are under going positive selection, as their nuclear counterpart.

HETEROGENEOUS GENOMIC DIFFERENTIATION IN MARINE THREESPINE STICKLEBACKS: ADAPTATION ALONG AN ENVIRONMENTAL GRADIENT

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Recent years have seen a shift in the way we view population structuring in organisms living in marine environments. Once thought of as panmictic and genetically homogenous, marine populations are now realized to have the potential to be subdivided on a finer geographic scale than previously appreciated. Descriptions of variation at genes whose functional roles are associated with specific selection regimes can offer insight towards the scale and degree of adaptive divergence among marine populations, but until recently this approach has traditionally been limited to very few loci. In our research, we use a genome-wide set of 140 microsatellite markers that are within (or close to) genes that have exhibited transcriptional responses to specific environmental conditions, in order to explore the patterns of adaptive population divergence and heterogeneous genomic differentiation among marine threespine sticklebacks (*Gasterosteus aculeatus*) in Northern European seas. Using 20 of these markers and a much denser sampling scheme, we then further explored adaptive divergence within the environmentally heterogeneous Baltic Sea in relation to the steep salinity and thermal gradients. The results of our candidate genome-scans provide support for the emerging view that, in spite of the high degree of physical connectivity in marine environments, there is a great deal of adaptive divergence among marine populations that is not apparent when neutral loci are analyzed.

SYMPATRIC SPECIATION IN TWO WILD RICE SPECIES: INSIGHTS FROM GENOME-WIDE SNPS AND EXPRESSION PATTERNS

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The genetic basis of speciation and adaptation is a fundamental question in evolutionary biology and remains largely unknown. Two wild rice species (Oryza nivara and O. rufipogon) are sympatric but distinct morphologically and ecologically. Thus they are typically incipient species and provide a unique opportunity to study ecological speciation at the genome scale. Here we investigate genomewide variation responsible for speciation between the two species using next-generation sequencing technology. First, we obtained 5.3 million SNPs based on the re-sequencing data with an average 2-10x coverage for 11 individuals from each of the two species. We located and characterized 126 genomic regions or islands that are significantly differentiated between the two species. These speciation islands located across all 12 rice chromosomes, encompassed 30.5 M (7.98%) of the genome and contained 2308 predicted genes. These candidate speciation genes involve mainly transcription factors, regulation of gene expression, flower development, pollination, and reproduction. Second, we used RNA-seq to measure gene expression in three different tissues of the two species. We obtained 15390, 18938, and 19311 expressed genes for flag leaves, booting panicle, and flowering panicle, respectively. Of them, 3.1-5.4% of expressed genes have divergent expression (DE) between the two species, with the number of under-expressed genes being slightly more than that of over-expressed genes in O. nivara for all three tissues. The GO annotations indicated that the DE genes involved the pollination, reproduction, growth and development, response to stress, secondary metabolic process, etc. These results demonstrate that both coding sequence variation and regulatory changes contribute to adaptive differentiation of species. This study provides new insights into the genetic mechanisms underlying the differentiation and adaption of two wild rice species and plant speciation in general.

STRONG SELECTION ON MITOCHONDRIA-TARGETING PROTEINS ACROSS SECONDARY CONTACT ZONES IN MACOMA BALTHICA

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Little is known on the genes and gene interactions involved in inter-population genetic divergence in the context of secondary contact. The marine bivalve *Macoma balthica* has a complex biogeographic history in the NE Atlantic, punctuated by multiple events of trans-arctic invasions and the establishment of secondary contact zones. We used transcriptome-wide DNA sequence data to explore patterns of divergence and selection across divergent *M. balthica* populations, with emphasis on genes involved in mitochondrial processes. Analysis of non-synonymous to synonymous mutation (dN/dS)ratios revealed that up to 5% and 45% of the 752 tested genes were under positive and negative selection (respectively), with genes involved in mitochondrial energy production detected in both categories. Over 87% of genes coding for mitochondrial-targeting proteins were under negative selection. We evaluated potential adaptive protein evolution of the ATP synthase gamma subunit, because strong population differentiation was detected for this gene. A sliding window analysis of dN/dS revealed that this gene, while under strong negative selection overall (dN/dS = 0.08), showed strong positive selection (dN/dS > 2) along a 46-residue fragment (15% of the sequence). This area contains peptide-binding sites that may interact with the epsilon subunit (F1) and the mitochondrialencoded F0 portion of the ATP synthase F0/F1 complex. Gene ontology terms associated with the mitochondrial machinery and multiple binding activities (including ATP binding) are enriched across populations, many of these terms being also enriched across divergent populations of the copepod Tigriopus californicus, a recognized model for the study of early stages of speciation in which mitonuclear incompabilities were detected. There is therefore accumulating evidence suggesting that mitochondrial-targeting proteins and their interactions play a role in maintaining population divergence in *M. balthica*.

GENOMIC SIGNATURES OF REPRODUCTIVE ISOLATION IN THE WHITE STICKLEBACK

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Knowledge of the genetic architecture of reproductive isolation is key to an integrated understanding of speciation. Species that diverged with gene flow (or exchange genes currently) allow for this architecture to be probed directly via gene flow mapping. This method is based on the observation that genomic regions involved in reproductive isolation resist migration (i.e. are more differentiated) more than other regions, which flow freely across species boundaries. Here, I explore the genetic architecture of reproductive isolation in a novel stickleback system: the white stickleback. White sticklebacks are sympatric morphs of the Atlantic marine threespine stickleback ("common" sticklebacks), found in Nova Scotia, Canada. Previous work has established that male white stickleback display brilliant white nuptial coloration, do not perform post-fertilization parental care and nest exclusively in filamentous algae. In spite of these differences, previous genetic data suggest that gene flow between white and common sticklebacks is likely ongoing. Here, we take advantage of this fact to probe the genetic architecture of reproductive isolation between white and common sticklebacks via gene flow mapping. Using the recently developed technique of Genotyping By Sequencing (GBS), we generated a SNP dataset containing thousands of markers. We used this to infer the demographic history and evolutionary relationship between white and common sticklebacks. We then performed genome-scans of population differentiation and signatures of natural selection in the two morphs, and identified regions likely involved in reproductive isolation and divergent adaptation between the two types. We compared the findings to other stickleback systems, and attempted to identify parallels between the two. Overall, our findings paint a complex picture of the interplay between adaptation and reproductive isolation in this novel system.

A POPULATION GENETIC MODEL OF ALLOPOLYPLOID SPECIATION

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Allotetraploid speciation, i.e. the generation of a hybrid tetraploid species from two diploid species, and the long-term evolution of tetraploid populations and species are important in plants. A population genetic model to infer population genetic parameters of tetraploid populations from data of the progenitor and descendent species is presented. The model links different time-scales: allopolyploid hybrid speciation is presumably rare and probably results in a population bottleneck, i.e., a founder event. Immediately after allopolyploid speciation, selection must be intense, as hybrids of early generations are usually relatively unfit but often evolve to out-compete their parental species and spread to occupy new ecological niches. Later, the accumulation of mutations may render one or the other homeologous copy of a gene dysfunctional and recombination or gene conversion may lead to mixing of two homeologous gene copies. These latter processes are relatively slow. In a first attempt, we develop a population genetic model to capture key parameters and apply this model to a data set of nuclear genes of two yarrow species, Achillea alpina-4x and A. wilsoniana-4x, that arose by allotetraploidization from the diploid progenitors, Achillea acuminata-2x and A. asiatica-2x.

INFERENCE OF SELECTION USING EXTENDED HAPLOTYPE HOMOZYGOSITY

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The fast-growing amount of genome-wide polymorphism data available has led to considerable efforts for developing methods to detect the footprints of natural selection at the molecular level. Finding regions under selection is one of the first steps to understand the process of adaptation and speciation. Our ability to detect selection at the molecular level depends critically on the type of data available and on the robustness of the methods to the underlying assumptions. Outlier methods are commonly used to infer selection. However, it has been shown that these methods have failed in some cases to clearly identify loci under weak selection and some markers in neutral regions of the genome have given strong signals of divergence (false positives). We study here the efficiency on simulated data of a recent method which allows to infer selection, which was firstly developed for human populations. This method, named iHS, is based on the concept that the action of selection on a given nucleotide will create a region of extended homozygosity around this nucleotide. Our results show that this method seems to only work when selection is strong and on a single locus, and fails to identify positions under selection in cases where selection acts on a quantitative trait coded by several loci.

DOES "COYNE'S RULE" APPLY TO AMPHIBIANS? INTROGRESSION OF SEX-LINKED AND AUTOSOMAL MARKERS ACROSS A TREE FROG HYBRID ZONE

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The large role of sex chromosomes in reproductive barriers between recently diverged species is one of the best-supported ideas in speciation research, and has been termed "Coyne's Rule". Most of this support, however, comes from studies of organisms with well-differentiated sex chromosomes such as fruit flies, mammals, and birds. In contrast to these groups, many fish and nearly all amphibians have homomorphic sex chromosomes, which contain a small sex-determining region and very large pseudo-autosomal regions. To date, little evidence is available to test how this dramatic difference in sex chromosome structure affects the genetic architecture of reproductive barriers. We genotyped 8 sex-linked and 16 autosomal microsatellites in frogs sampled along two transects across the hybrid zone between Hyla arborea and Hyla orientalis, and estimated cline width and position in order to test for differential introgression. Our results show no significant difference between introgression of sex-linked and autosomal markers, suggesting that the large effect of sex chromosomes on speciation may be limited to species with old and highly divergent sex chromosomes.

GENOME WIDE ASSOCIATION OF HIGH DIVERGENCE, TRANSMISSION RATIO DISTORTION AND QTL FOR MALE FERTILITY IN HYBRIDS BETWEEN ARABIDOPSIS LYRATA SUBSPECIES

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Crosses between evolutionarily diverged populations may result in sterile hybrids, which can eventually lead to speciation. In plants, early evolution of male sterility can be caused by incompatibilities between maternally and bi-parentally inherited genomes. Another phenomenon associated with high divergence of genomes is distorted transmission of gametes of F1 hybrids. Genome wide divergence data allow us to examine whether transmission ratio distortion and the regions affecting hybrid male fertility co-locate with islands of high divergence. We examined post pollination reproductive isolation between subspecies of the perennial outcrossing self-incompatible Lyrate rockcress (Arabidopsis lyrata) by crossing experiments between two highly diverged populations. We grew large reciprocal F2 progenies and three generations of backcrosses. F2 plants were genotyped for altogether 96 markers and the genomes of their parents were sequenced on Illumina platform. Transmission of most of the markers was distorted indicating incompatibilities between the diverged genomes. We observed cytoplasmic male sterility in almost one fourth of F2 and half of backcross hybrids in one of the reciprocal crosses. This asymmetrical hybrid incompatibility is due to sterility factors in one cytoplasm, for which the other population lacks effective fertility restorers. However, genotyping and genetic mapping revealed that only 60 % of the plants having the male sterile cytoplasm and lacking the corresponding restorers were phenotypically male sterile. We found that there is only one male fertility restorer locus that mapped to a 600 kb interval at the top of chromosome 2 in a region containing a cluster of pentatricopeptide repeat genes. At the same genomic region, there is also high sequence divergence between populations. We also compare genome wide transmission ratio distortion to divergence and discuss the role of cytoplasm and genomic conflict in incipient speciation.

EXTREMELY LOW GENETIC DIVERSITY AND POPULATION DIFFERENTIATION IN A SOCIAL INBREEDING SPIDER

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Mating systems strongly influence many aspects of species' biology, among them the level and structure of genetic variation. Evolution of sociality in spiders is associated with a transition from outcrossing to inbreeding mating system. All known social spiders (~23 species) are relatively young species that recently evolved regular inbreeding. Social spiders live and mate in colonies of highly related individuals with, while their solitary sister species disperse before mating. In addition they show highly female biased sex ratio (~8:1), reproductive skew, and are characterized by strong metapopulation dynamics. All these factors are predicted to affect their population genetic patterns towards low diversity and, depending on the strength of meta-population dynamics, high among-population variation due to reduced gene flow among populations. *Stegodyphus sarasinorum* is a social spider distributed across the Indian sub-continent and adjacent countries. Spiders were sampled over more than 3000 km from Sri Lanka in the south to Himalaya in the north with denser sampling in central India, and 17 coding loci were sequenced in all individuals. As predicted, the diversity was found to be very low, both at the population and the species level. Interestingly, at this level of sequencing no population differentiation could be identified, suggesting a strong homogenization of genetic variation over extremely large areas caused by meta-population dynamics.

DIVERGENCE BETWEEN LAKE AND STREAM POPULATIONS IN AN EAST AFRICAN CICHLID FISH

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Integrative studies of species that occur along an environmental gradient provide important insights into ecological speciation and serve as prime examples for the presence of a speciation continuum. The cichlid species Astatotilapia burtoni occurs in lacustrine environment as well as in the surrounding rivers of Lake Tanganyika (LT), offering the possibility to study a lake-stream environmental gradient in a member of a large cichlid adaptive radiation in East Africa. We have established phylogeographic relationships and assessed the population structure in A. burtoni from the southern LT drainage using neutral nuclear (microsatellite) and mtDNA markers. We detect an unexpectedly high genetic diversity in A. burtoni, exceeding – at least in mtDNA – the diversity of the entire cichlid species flock of Lake Victoria, and a relatively complex phylogeographic pattern. Next, we have examined morphological differences among these populations by analyzing body shape. Based on these results, we focused on four lake-stream systems in detail. We here find that stream fish show a more inferior mouth, a more streamlined body, and shorter gill-rakers compared to lake fish, shifts presumably associated with differential foraging styles. Current work is now evaluating this hypothesis directly, and includes high-density SNP marker data generated by RADseq for population genomic analyses.

THE GENETIC ARCHITECTURE OF DIVERGENT POLLINATOR ATTRACTION IN SEXUALLY DECEPTIVE ORCHIDS

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Sexually deceptive orchids offer a fascinating system for studying ecological speciation, where species divergence is driven by adaptations to different specific pollinators. Here, differences in floral odour bouquets are largely responsible for pollinator-mediated reproductive isolation. Using comparative RNA-Seq analysis in combination with ecological and molecular approaches, we investigate the genetic architecture and molecular mechanisms that underlie traits relevant for specific pollinator attraction. Comparisons of sequence polymorphisms and expression levels allow us to pinpoint genes likely under selection that directly contribute to divergent phenotypes associated with differences in plant fitness. Likewise, we can now address the question of how interactions among genes can constrain of facilitate the divergence of lineages.

A YOUNG NEO-SEX CHROMOSOME IS THE SITE OF DIVERGENCE BETWEEN INCIPIENT STICKLEBACK SPECIES

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Sex chromosomes turn over rapidly in some taxonomic groups, where closely related species have different sex chromosomes. However, little is known about the functional roles of sex chromosome turnover in phenotypic diversification and speciation. We use a sympatric pair of threespine sticklebacks to understand the roles of sex chromosome turnover in phenotypic divergence and speciation. Previously, we found that the Japan Sea sticklebacks have a neo-sex chromosome system resulting from a fusion between an ancestral Y chromosome and an autosome, while sympatric Pacific Ocean sticklebacks have a XY sex chromosome system. Furthermore, we demonstrated that the Japan Sea neo-X chromosome plays an important role in phenotypic divergence and reproductive isolation between these sympatric species. Here, we conducted additional QTL mapping of other morphological and behavioral traits to confirm that both old-X and neo-X chromosomes have significant QTL important for morphological divergence, while the old-X, but not the neo-X chromosome, contributed to hybrid courtship abnormality. Next, we conducted whole genome sequencing of these two sticklebacks to find that the Japan Sea neo-Y chromosome has no large-scale deletion, but some regions on the neo-Y have started to accumulate deleterious mutations. Importantly, significant QTL on the neo-X chromosome were located at regions even outside the regions where deleterious mutations accumulate. Furthermore, comparison of genome sequences between these two species revealed that the neo-X chromosome showed faster protein evolution than autosomal genes. Thus, a young neo-sex chromosome is the site of divergence between incipient species.

QUANTITATIVE TRANSCRIPTOME PATTERNS IN THE ALLOPOLYPLOID S. ALBURNOIDES COMPLEX

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Squalius alburnoides is an allopolyploid fish complex characterized by a constant switch of genome compositions and ploidy level changes in consecutive generations. This promotes recurring situations of potential genomic shock that S. alburnoides successfully overcomes. Previously, in a study involving only few genes, it was seen that the transcription levels in diploid and triploid S. alburnoides were similar. In this work we assessed if this described compensation mechanism that reduces the transcript levels of some genes to the diploid state, occurs globaly in the transcriptome of this natural occurring allotriploids. Also, we want to determine if there is a genome specific dominance or a similar allelic contribution to the overall expression in these hybrids. We did "RNA-seq Illumina sequencing to perform a first comparative full transcriptomic analysis of the most frequent S. alburnoides complex forms (PA; PAA; AA and PP). For the first time, high-throughput gene expression levels between diploid and triploid forms of a hybrid vertebrate organism were determined and compared. Also, polymorphisms between genomes (P and A) were identified and the allele specific gene expression (ASGE) was estimated in the hybrids. The results indicate that there is a clear leaning towards positive nonaditive gene regulation following interspecific hybridization, and a global tendency to negative nonadditive expression in the triploid hybrid. Concerning ASGE, results show that there is no massive preferential allele/genome specific gene copy silencing but there is a bias towards increased amount of P allele expression. We also explored in silico a possible interaction of miRNA's known to be up regulated in triploid S. alburnoides with identified genes with negative non additive expression in the same genomic composition. The results did not reveal a relevant involvement of miRNAs in the decay of specific mRNAs.

EFFICIENT BENCHTOP POPULATION GENOMICS AND AND HIGHLY POOLED POPULATION GENETIC SCREENING WITH RESTRICTION FRAGMENT SEQUENCING (RESTSEQ)

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We developed a new and efficient method (RESTseq) to employ benchtop next generation sequencing for efficient population genomic and genetic studies. In contrast to existing methods like RAD or GBS, this improved approach is a valuable alternative for a cost efficient, highly flexible and repeatable enrichment of DNA fragments from digested genomic DNA. With our method the sample preparation is fully scalable making it available for any platform including small scale Personal Genome sequencers like Ion Torrent. Easy adjustments to this method provide a very conveniant opportunity for sequencing highly pooled samples for population genetic studies aiming at only small numbers of SNP markers. Thus it can be used for large scale (high numbers of markers or individuals) SNP genotyping in model and non-model organisms. We demonstrate the validity and reproducability of our approach by comparing two honeybee samples at genomic level with genotyping several thousand SNP markers as well as with a pooled sample of a non-model stingless bee aiming at less than 1000 SNPs per individual for a population genetic study.

IS HYBRID SPECIATION DRIVEN BY STOCHASTIC OR DETERMINISTIC PROCESSES?

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Natural hybridization may have a multitude of consequences for biological diversity. One possible outcome is hybrid speciation. Whether this mode of speciation is primarily driven by stochastic or deterministic processes remains an open question. Recently, it was shown that the Italian sparrow (Passer italiae) is a hybrid species formed by interbreeding between the house (P. domesticus) and Spanish sparrow (*P. hispaniolensis*). Today, the parent taxa occur sympatrically over large parts of their breeding range. This allows for a rare opportunity to recapitulate the processes that were involved in the formation of this hybrid species. Previous studies have shown that the Italian sparrow is fixed for house sparrow mitochondria while mito-nuclear incompatibilities appear to act as strong reproductive barriers against its other parent species, the Spanish sparrow. Further, sex-linked genes appear to be involved in reproductive barriers between the hybrid species and both parents. Studying a sympatric population of house and Spanish sparrows on the Iberian Peninsula, we recover a similar pattern. We find strong asymmetry in introgression patterns between the two parental species as well as asymmetry in introgression patterns among genomic regions. All but one of the 60 admixed individuals out of a total of 292 sampled birds had house sparrow mitochondria. Moreover, sex-linked markers in general, and those with known mitochondrial function in particular, introgressed at much lower rates into opposite mitochondrial-genetic background than did autosomal markers. These striking similarities in introgression patterns across parallel systems suggest that the formation of this hybrid species was driven by deterministic rather than by stochastic processes.

RATE OF MOLECULAR EVOLUTION FOR GENES IN REGIONS WITH LIMITED GENE FLOW ACROSS THE HOUSE MOUSE HYBRID ZONE

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Two (sub)species of house mouse (*Mus mus musculus/M. m. domesticus*) form a hybrid zone across Europe maintained by dispersal of individuals and selection against hybrids. Multiple regions on the X chromosome and autosomes have been previously shown to have reduced introgression across the hybrid zone. In this study we explore rate of molecular evolution inferred from rate of nonsynonymous and synonymous substitutions (dN/dS) in these regions to identify fast evolving genes as possible candidates involved in reproductive isolation. The nonsynonymous and synonymous substitutions were retrieved from a comparison of the reference mouse genome, mouse strains representing *M. m. musculus* (PWK/Ph) and *M. m. domesticus* (WSB/Ei) (sub)species and rat. Based on dN/dS ratios we have identifieda suite of candidate reproductive isolating genes.

SPECIATION HISTORY OF THE ATLANTIC EELS

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Allopatric speciation has generally been considered the most prominent process for the evolution of new species. Although evidence for sympatric speciation has increased recently it is still controversial to what degree it occurs in nature. Hence, it is generally accepted that allopatric speciation should be the null hypothesis against which sympatric speciation is tested. Marine environments often have no obvious geographic barriers, which makes sympatric speciation a likely scenario. However, only few marine organisms have the characteristics that make them good candidates for testing mode of speciation. The European and American eel (Anguilla anguilla and A. rostrata) are panmictic species that spawn in the Sargasso Sea. Their spawning area and time overlap considerably. Hybrids are rarely observed, indicating reproductive barriers. In this study we conduct whole mitochondrial genome and RAD sequencing to investigate the speciation history of the two Atlantic species of eel. Using coalescence based analyses we estimate divergence time to test the hypothesis that speciation occurred long time ago in sympatry. As a result of continental drift the distance between the spawning grounds increased, causing selection on e.g. duration of the larval phase. The alternative hypothesis concerns speciation in allopatry during the Pleistocene. Glaciations significantly decreased the amount of available habitats in coastal regions and freshwater bodies. Hence, population size is likely to have undergone a severe decrease. The smaller population sizes of European and American populations would result in reduced overlap of spawning area leading to reproductive isolation. The impact of the last glaciation on effective population size is assessed by conducting Bayesian skyline plots based on the mtDNA. Finally, patterns of genome-wide selection between the two species are analyzed using RAD sequence data in order to detect genes and genomic regions involved in speciation.

23. Genomic Islands: Their Role in Adaptation and Speciation

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ECOLOGICALLY DRIVEN MORPHOLOGICAL AND GENETIC DIVERGENCE BETWEEN SHELTERED AND EXPOSED POPULATIONS OF THE MARINE SNAIL *LITTORINA FABALIS*

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It is well established that ecology plays a critical role in the process of population divergence and ultimately in speciation. Nonetheless, our understanding of how it occurs is still scarce. Characterized by abrupt changes in the environmental conditions over a narrow spatial scale, the marine intertidal is an ideal system to provide valuable insights into the field of ecological speciation. In our study, we focus on North Atlantic populations of the flat periwinkle *Littorina fabalis* to explore to what extent adaptation and genetic divergence are promoted by contrasting wave exposure intensities and other associated factors. We analysed shell morphology and performed a genomic scan based on AFLP loci to estimate divergence between sheltered and exposed populations of *L. fabalis* at both micro (<40 Km) and macro-geographic (>1000 Km) scales. With this nested sampling approach, we aim to quantify phenotypic differentiation and to identify loci under disruptive selection among *L. fabalis* populations subject to different selective pressures in a repetitive manner along the species range. Doing so, we hope to shed light on the genetic mechanisms of ecotype evolution and ecological speciation, ultimately contributing to discern how repeatable (parallel) is divergent evolution in *L. fabalis*.

TESTING THE LINK BETWEEN HETEROCHROMATIN EVOLUTION AND SPECIATION IN STICKLEBACKS

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Genes that cause hybrid incompatibility between closely related species have not yet been identified in natural vertebrates. Stickleback fishes are a great model for identifying speciation genes, because there are multiple sympatric species pairs (i.e., pairs of incipient species that are sympatric, but are reproductively isolated). Our previous studies identified several QTLs of isolating barriers, including hybrid male sterility, between two sympatric incipient species of Japanese threespine stickleback fishes. Theoretical study suggests that heterochromatin-binding proteins can rapidly evolve in selfish manner and may affect the genetic incompatibility. Indeed, causal genes of hybrid male sterility in Drosophila and mice are rapidly evolving genes involved in heterochromatin regulation. To test the link between heterochromatin evolution and speciation in natural vertebrate systems, we determined the whole genome sequences of these two stickleback species and analyzed the sequence differences in the QTL regions in order to identify candidate speciation genes. Within the QTL regions of hybrid male sterility, we found two chromatin-binding domain containing genes with high Ka/Ks rate. Currently, we are analyzing the function of these genes in vitro using protein-protein interaction assays and in vivo using the transgenic sticklebacks.

CONSTANS IS HIGHLY PRONE TO NATURAL SELECTION IN ARABIDOPSIS LYRATA

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We examined the evolution and diversity of genes related to flowering time, an important adaptive trait in plants, and a set of unrelated reference genes in the perennial Arabidopsis lyrata in several populations in Europe and North America. Among the 38 loci, CONSTANS, one of the central flowering time genes, was the only which had experienced species-wide adaptive evolution when compared to A. thaliana, as shown by multilocus MK analysis. This was consistent with earlier result of selection on this gene in A. thaliana (Bustamante et al. 2002), and of the rapid evolution of this gene in Brassicaceae (Lagercranz & Axelsson 2000). The CONSTANS gene also was highly differentiated between the widespread A. lyrata populations (Fst=0.78). Further, an association analysis suggested a role in governing fitness in an experimental field site at the University of Oulu. Thus, we found CONSTANS gene to be adaptively important in all studied time levels of natural selection. Likely selection on flowering time, may often target the CONSTANS gene.

GENETIC CHANGES UNDERLYING HOST PLANT RESISTANCE IN DROSOPHILA SECHELLIA

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The genetic basis of adaptation is a major question in evolutionary biology. In particular, ecological specialization can result from rapid host shifts and strong selective forces. Although most Drosophila species are generalists, D. sechellia a species endemic to the Seychelles archipelago is one such specialist species. D. sechellia feeds and breed almost exclusively on Morinda citrifolia, and its fruit commonly known as noni. *M. citrifolia* produces several volatile compounds which are toxic for most arthropod species and all closely related *Drosophila* species. Nonetheless, *D. sechellia* has evolved resistance and host preferences that include stimulation of egg production, oviposition site preference and chemotaxis. To understand how this suite of host specialization traits evolved, it is important to dissect the underlying genetic basis of these phenotypes. QTL mapping is an important tool for the study of such complex traits. In order to detect and localize loci that influence resistance to M. Citrifolia, we use bulk segregant mapping approaches, based on backcrosses between D. sechellia and its sister species *D. simulans* (which is sensitive to Noni) and use by deep sequencing to identify QTL regions. We combine this approach with genome scans, based on light coverage sequencing of over 250 isofemale lines of *D*. Sechellia and *D*. Simulans derived from recent field collections in the Seychelles. This approach allows us to unravel the genetic characteristics that allow *D. sechellia* to contend with the toxic compounds that are otherwise lethal to close relatives.

FROM ECOLOGICAL SPECIALIZATION TO CRYPTIC SPECIATION IN THE PARASITIC WASP COTESIA SESAMIAE

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Cotesia species (Hymenoptera, Braconidae) are endoparasitoids of Lepidoptera larvae. Their virulence is mediated by a polyDNA symbiotic virus (Bracoviridae) of which genome is integrated to the wasp chromosomes. In Cotesia sesamiae Cameron it has been shown that host pressure exerts positive selection on the virulence gene CrV1, and that the evolution of the bracovirus genome is associated to cases of host specialization (Gauthier et al., this meeting). Here we studied the species status of *C*. sesamiae host races. It attacks a wide range of stemborer larvae, including pests of maize and sorghum in sub-saharan Africa. Our study was based on a geographically and ecologically diverse sampling. A phylogenetic analysis conducted on seven-gene concatenated tree revealed two major clades. One has a diversified ecological niche and gathers host races associated with distinct CrV1 allelic variants, supporting the hypothesis that conserved gene flow between populations maintains their ability to shift between hosts. In contrary, the second clade is invariably associated with one host species, Sesamia nonagrioides, on one host plant, Typha domingiensis. It harbors a unique CrV1 allele, namely Cs Snona, not present in the first clade. Further investigations allowed us to give the status of a new species, labelled C. typhae, to this lineage. (i) Cross mating experiments revealed reproductive isolation from two populations of the other lineage, as well as from *C. flavipes*, a sister species of *C. sesamiae*; (ii) Experimental analysis of parasitic success showed that C. typhae was the only lineage virulent against S. nonagrioides. Hence, C. typhae constitutes a new species in the C. flavipes complex, potentially resulting from sympatric speciation. It is characterized by reliable ecological niche and molecular markers. It represents a potential biological control agent for southern Europe where S. *nonagrioides* is a major pest of maize crops.

Symposium

24. Unifying Paleobiologiccal and Comparative Perspectives on Character Evolution

22 and 23 August



Program

Thursday 22 August Session(s): 9 Friday 23 August

Session(s): 10

Organisers: Lee-Hsiang Liow and Thomas F. Hansen

Invited speakers: Gene Hunt and Folmer Bokma

Description:

It is no longer debated that the fossil record is necessary to inform us about the history of life, yet the integration of data and perspectives using fossils and comparative data in understanding evolution is far from mature. This symposium gathers researchers straddling the realms of the extinct and the extant to explore how we can better understand evolutionary processes especially on time scales common to palaeobiological and phylogenetic comparative studies, using character evolution as a focal point.

24. Unifying Paleobiological and Comparative Perspectives on Character Evolution

D22SY24IT10:30R7

THE FOSSIL RECORD OF LINEAGE EVOLUTION

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The fossil record captures the only direct observations we have of evolutionary changes within species over substantial spans of time, and yet it has long been controversial as to how best to interpret such observations. In this talk, I review some of the important accumulated findings from paleontological research about the nature of evolutionary changes within fossil species. Specifically, I will address the frequency of different modes or patterns of evolutionary change, the tempo of these changes, and the implications of these for understanding evolutionary processes in the fossil record. In addition, I will discuss case studies that: (1) estimate the importance of anagenetic (within-lineage) versus cladogenetic (at speciation) changes in phenotypic traits, and (2) test hypotheses about specific causal drivers of phenotypic change in fossil species.

D23SY24IT10:30R7

TEMPO AND MODE IN EVOLUTION OF COMPLEX ORGANISMS.

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Most evolutionary biologists appear to believe that populations and species are well adapted to their environment, as the result of continuous action of natural selection on all their traits. In complex organisms, however, many traits do not experience the outside environment: their environment consists of other traits. This implies that fitness is not determined exclusively by how well traits fit the external environment, but also by how well traits are adapted to each other. I will discuss whether the existence of co-adapted trait complexes may determine the tempo and mode in the evolution of their component traits, and discuss how we could distinguish the extrinsic and intrinsic components of fitness in phylogenetic and experimental studies.

D22SY24RT11:18R7

SEXUALLY SELECTED TRAITS IN THE FOSSIL RECORD

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Sexual selection is one of the most important driving forces behind the diversity that we see in modern biological systems, and it is increasingly being recognised that sexual selection probably plays an important role in large-scale processes such as speciation, extinction and adaptation. It has, however, been largely ignored by many palaeontologists despite the large number of high-profile examples of apparent sexually selected features in the fossil record. There are a number of reasons why this is so, including the difficulty of assigning sex to most fossils, the problem that the more extreme sexual dimorphs are likely to be described as different species, the existence of sexual dimorphs that have arisen because of natural rather than sexual selection and the sheer variety of sexually selected traits, which makes it very hard to identify specific morphological features that can be used to distinguish sexually selected traits from others. These problems are not insurmountable and with careful thought and well-planned hypothesis testing sexually selected traits can be distinguished from others in the fossil record, opening up opportunities for firstly a better understanding of the biology and behaviour of extinct animals and secondly for the use of the fossil record in comparative studies to test ideas about the role of sexual selection in driving processes like adaptation and speciation.

D22SY24RT11:42R7

DECOUPLING OF TAXONOMIC DIVERSITY AND MORPHOLOGICAL DISPARITY DURING DECLINE OF THE CAMBRIAN TRILOBITE FAMILY PTEROCEPHALIIDAE

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Though discordance between taxonomic diversity and morphological disparity is common, little is known about the underlying dynamics that drive this decoupling. Early in the history of the Cambrian trilobite family Pterocephaliidae, there was an increase in taxonomic diversity and morphological diversity. As taxonomic diversity declined in the later history of the clade, range of variation stayed high, and disparity continued to increase. However, per-branch rates of morphological evolution estimated from a recent phylogeny decreased with time. Neither within-trait nor within-species variation increased or decreased, suggesting that the declining rates of morphological evolution were more likely related to ecological opportunity or niche partitioning rather than increasing intrinsic constraints. This is further supported by evidence for increased biofacies associations throughout the time period. Thus the high disparity seen at low taxonomic diversity late in the history of this clade was due to extinction—either random or targeting mean forms—rather than increased rates of morphological evolution. Furthermore, patterns seen at higher taxonomic scales do not necessarily reflect patterns at lower levels or within an explicit phylogenetic framework. These results emphasize the importance of considering both extinction and phylogenetic context when comparing different types of diversity and seeking explanations for conflicting patterns. This pattern also provides a scenario that could account for instances of low taxonomic diversity but high morphological disparity in modern groups.

D23SY24RT11:18R7

150 MILLION YEARS OF MORPHOLOGICAL AND FUNCTIONAL EVOLUTION IN NEOPTERYGIAN FISH: IMPLICATIONS FOR NOTIONS OF TELEOST SUPERIORITY

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Teleost fishes are the dominant group of extant vertebrates; they include approximately 29,000 species, assume a bewildering array of morphologies, and have come to occupy nearly every aquatic environment. In dramatic contrast, their holostean sister group comprises a mere 8 living species, all of which are restricted to the freshwaters of eastern North America. It is this pattern of extreme dissimilarity, gleaned from living taxa alone, which has provided the basis for assertions of teleost 'superiority' and fuelled a series of evolutionary scenarios. However, the fossil record indicates that these groups arose in the Permian, and so around 250 million years of diversification has been largely excluded from the debate. By reconstructing the historical diversity trajectories for these groups, we can establish the pattern by which teleosts came to dominate. We quantify the morphological and functional diversity holosteans and teleosts across 150 million years of the Mesozoic (representing 70% of neopterygian history). Contrary to the pattern seen in extant taxa, holosteans appear morphologically and functionally superior over teleosts in the Triassic. However, teleost morphological and functional diversity increases in the Jurassic to the point that they overtake holosteans by the Lower Cretaceous. Bayesian analysis of evolutionary rates only weakly supports the notion that teleosts exhibit higher rates of morphological change than their holostean sister group.

D23SY24RT11:42R7

ALLOMETRIC CONSTRAINTS ON ADAPTIVE RADIATION IN STICKLEBACKS

<u>Kjetil L Voje</u>¹, Anna B Mazzarella¹, Thomas F Hansen¹, Kjartan Østbye¹, Tom Klepaker³, Arthur Bass¹, Anders Herland¹, Kim M Bærum², Finn Gregersen⁴, Asbjørn Vøllestad¹

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The morphological differences between the marine ancestor and the descendant freshwater populations of threespine sticklebacks constitute a well-studied example of a phenotypic radiation. However the exact selective agents that drive these changes are not yet fully understood. We present a comparative study across 74 freshwater populations of threespine sticklebacks in Norway to test whether evolutionary changes in stickleback morphology can be explained as adaptations to lake characteristics thought to reflect different habitats and feeding niches. Only weak indications of adaptation were found, and the rates of adaptation varied from immediate to more constrained evolution among traits. Instead, populations have diversified in phenotypic directions predictable from allometric scaling relationships. This indicates that evolutionary constraints may have played a role in structuring phenotypic variation across freshwater populations of stickleback.

POSTERS

D23SY24PS0186

EVOLUTIONARY ECOLOGY AND EXTINCT SPECIES: HOW DO FOSSILS CHANGE OUR INTERPRETATION OF PRESENT-DAY BIODIVERSITY PATTERNS? USING PRIMATES AS AN EXAMPLE

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Studying events in the evolutionary history of a taxonomic group, such as adaptive radiation or extinction, requires a fine-scale and accurate resolution of their relationships through time. To achieve that, most scientists would agree that information about both extant and extinct species is needed. However few efforts have been made to combine extant and extinct species in the same phylogenetic trees, instead phylogenetic trees usually contain only extant species. Because the vast majority of species in a lineage will be represented by extinct species, studies focusing on extant species contain less than 0.1% of the lineage's species richness. In some clades, ignoring extinct species may also obscure the true evolutionary history, species richness (i.e. Proboscidea), biogeography (i.e. Tinamiforms) or ecological diversity (i.e. Crocodilomorphs). Thus including the fossil record in these kinds of studies is essential to fully understand the evolutionary history of lineages. In this study, we constructed a new primate phylogeny based on Ronquist et al.'s (2012) Total Evidence Method. We used 73 genes from over 340 extant primates and approximately 350 morphological characters from over 250 species (half of which were extinct species). We applied phylogenetic comparative methods to investigate evolution of body mass variation through time in Primates. We show that taking account of extinct species morphological traits such as body mass affects the estimation of evolutionary rates especially among Lemuriforms (Strepsirhinni) where the evolutionary rates are highly increased.

D23SY24PS0195

ADAPTIVE RADIATIONS IN ALPHA AND BETA NICHE DIMENSIONS

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Adaptive radiations are interesting and important, and probably more than just a sequence of speciations. Recently, there has been an increased interest in the more exact sequence of events, how a clade develops from its first ancestral population to a group of closely related species, adapted to different niches in the landscape in which they occur. Some traits seem to be conserved through evolutionary history, whereas some are more labile, with little phylogenetic signal. The concepts of alpha and beta niches, corresponding to within- and between-habitat selection, has made it possible it hypothesize that one set of traits diverge sooner than others. However, data points in different directions. Moreover, theory is essentially lacking. I here present a first attempt to disentangle the mechanisms underlying the sequence of trait divergence. A first, tentative, prediction is that traits with the strongest trade-off will diverge first. Model simulations show that this pattern is surprisingly robust to the level of dispersal between habitat types. They also show that there is more to learn about the ecological mechanisms driving adaptive radiations.

D23SY24PS0311

DEVELOPMENTAL MECHANISMS AND EVOLUTION OF SHELL COILING IN GASTROPODS

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Various shapes of gastropod shells have evolved ever since the Cambrian. Although theoretical analyses of shell morphogenesis exist, the molecular basis of shell development remains unclear. In order to understand developmental mechanisms and evolution of shell formation in gastropods, we focused on the decapentaplegic (dpp) gene that is expressed around the shell gland. We compared expression patterns of dpp in the shell gland and the mantle tissues at various developmental stages between coiled and non-coiled shelled gastropods. In the limpets, Patella vulgata and Nipponacmea fuscoviridis, dpp showed symmetric expression patterns throughout ontogeny. On the other hand, in the coiled shelled snail Lymnaea stagnalis, dpp showed asymmetric expression and mirror image patterns between the dextral and sinistral lineages. Moreover, we analyzed the functions of dpp using the Dpp signal inhibitor dorsomorphin. When the embryos were treated with Dpp signal inhibitor at the trochophore or veliger stage after the shell gland formation, juvenile shells grew to show a cone-like form rather than a normal coiled form. These results suggest that the shell coiling is controlled by dpp, and that the loss of coiling in Patellogastropoda was caused by loss of the asymmetric expression of dpp in the shell gland at the trochophore stage, which lead to symmetric dpp expression at the veliger and adult stages.

D23SY24PS0845

REASSESSING THE TEMPORAL EVOLUTION OF SALAMANDRIDS: THE INTEGRATIVE EVALUATION OF TIME CONSTRAINTS CHALLENGES CURRENT HYPOTHESES

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Two main types of time constraints can be implemented to perform divergence time estimation analyses: paleontological and paleogeographic. The first ones are set using fossils, which usually are placed in nodes of the phylogenetic trees corresponding to the common ancestor to the members of particular extant clades. Those usually allow only setting minimum dates for particular nodes on the phylogeny, which are set as priors in the analyses. Maximum dates for most clades are not available or are questionable, and the interpretation of fossils may derive in different placements in the tree, which can affect divergence time estimations and reconstruction of the evolution of characters. Paleogeographic constraints correspond to historical events that may have caused splits of ancestral lineages, in which two sister taxa occur on both sides of a barrier. However, these cladogenetic splits assigned to particular paleogeographic events may have been caused by other factors, even in different time periods that do not overlap with the assumed paleogeographic ones. Moreover, dispersal across potential barriers have been shown to be more common than previously thought. Here we analyzed and tested every time constraint available for salamanders of the family Salamandridae, including the effect of the molecular dataset and priors on divergence time reconstruction. Our results show important effects of all these factors that challenge current placement of some fossils as well some historical biogeographic events, affecting also our interpretation of how and when some characters have evolved.

D23SY24PS0848

INSIGHTS ON NEUROANATOMY EVOLUTION OF EARLY MAMMALIAN ANCESTORS FROM A NEW PERMIAN DICYNODONT FROM MOZAMBIQUE

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A nearly complete skull and mandible, with a series of 19 articulated dorsal, sacral and tail vertebrae, ribs, ilia, partial pubis and femur (ML1620) was collected from the Late Permian Karoo sediments, Metangula Graben, northern Mozambique (Niassa Province), Cádzi Formation. The 3D visualization of the internal cranial bones, via µ-CT, combined with a phylogenetic analysis demonstrates a set of characters shared with Emydopoidea, a restricted clade of small-sized dicynodonts. Brain morphology and location of main cranial nerves is visualized by a virtual 3D model of the fossilized endocranial cavity. The brain is delimited by the basioccipital, exoccipital, supraoccipital and opisthotic posteriorly; by the postparietal, parietal, preparietal, and frontal dorsally; prootic and epipterygoid laterally; parasphenoid and basisphenoid ventrally; the olfactory tracts are delimited by the ethmoid ventrally. The brain is narrow and the cerebellum is broader than forebrain, resembling the condition of other non-mammalian therapsids. The orbits are located far anteriorly relative to the olfactory bulbs. The olfactory bulbs are separated from the cerebrum by a long olfactory tract. The anatomy of osseous labyrinth, pristinely preserved, is compared with the scarce data published on synapsid inner ear anatomy. These are the first detailed brain and auditory apparatus model of dicynodonts based on µ-CT. The preliminary observations indicate that there is a conservative reptilian-grade brain morphology governing the anatomy of non-mammalian synapsids.

D23SY24PS0900

GIANTS FROM DWARFS: SUPPORT FOR SYMPATRIC PROCESSES OF SIZE DIVERGENCE IN AUSTROLEBIAS SOUTH-AMERICAN ANNUAL KILLIFISH

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To determine the plausibility of sympatric speciation, we need to determine its relative rate of occurrence across different speciation events. Here we focus on a small taxonomic group and a particular speciation mechanism which requires sympatry: the emergence of a large and small species pair where the large cannibalizes the small. This scenario has been named "giant-dwarf" diversification. A comparative analysis of body size measures of Austrolebias South-American annual killifish suggests that species evolve towards one of three size optima. Species evolving towards the largest optimal size appeared at least three times from small in the Austrolebias genus. The first large ancestral species per event appeared in a trait change with relatively high speed in all three cases, consistent with expectations of cannibalism evolution. A comparative analysis of lower jaw length, a proxy for the level of specialization in piscivory suggests that in one clade of large species trait values indicate a very weak or no specialization towards piscivory, and that in the two other clades species are selected towards two optima with relatively large jaw lengths. By means of a reconstruction of ancestral species ranges we show that speciation events leading to a large and smaller species pair were sympatric with a large likelihood. For the clade of large species with little specialization, the probability that speciation was non-sympatric is largest among the three events. Conditional on the data we analysed, one can therefore conclude that giant-dwarf speciation by cannibalism most probably occurred twice in Austrolebias and that a third appearance of large species in the genus likely occurred by other selective or non-selective processes.

D23SY24PS1208

NEW APPROACH TO OLD VERTEBRAE – A MORPHOMETRIC COMPARISON OF MIOCENE AND EXTANT SNAKES

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Comparative morphological study of fossil and recent snakes is of key interest for taxonomic identification, paleobiological reconstructions and character evolution analyses. Snake fossil record is sparse, due to the low occurrence of fossilisation of delicate bone structures, especially in smaller species. The characters used in study of extant snake vertebrae are most often degraded in fossils protruding structures or exposed surfaces get broken off or eroded. The focus of this study was to explore the usability of structures less prone to damage, located near the centrum and at the base of the neural arch. This approach would allow the inclusion of a larger number of specimens, which could not be studied by the classical approach. We analyzed fossil snake vertebrae from a rich mid-Miocene assemblage at Vračević locality, in the vicinity of Belgrade, Serbia. We selected a sample of undistorted trunk vertebrae in good condition, referred to genera *Natrix*, *Vipera* and *Elaphe*. The fossil sample was compared to extant snake vertebrae, belonging to appropriate taxa. Three axial and four pairs of lateral landmarks (11 landmarks in total) have been identified on all the specimens and used for 2D geometric morphometric analysis. Bilateral acquisition addressed the problem of taphonomic deformation. Symmetric component of shape variation was used for the comparison of fossil vertebrae with extant samples. Fossil specimens had lower mean centroid size (CS), and higher variation in CS compared to recent ones. The range of fossil CS values was larger than the recent (the smallest and largest vertebrae were fossils). Principal component analysis (PCA) showed that the highest variation in shape was associated with centrum/neural arch height ratio. For further research, more comprehensive sampling is needed to rigorously evaluate the applicability of this approach.

D23SY24PS1276

HOW GOOD IS GOOD ENOUGH? AN INVESTIGATION OF TREE-BASED ANALYSES USING PALEONTOLOGICAL PHYLOGENIES

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Many tree-based methods now available for macroevolutionary analyses can include extinct taxa. Unfortunately, many of the groups for which we have abundant and diverse fossils have not been subjected to cladistic analysis. It is therefore important to understand the sensitivity of tree-based analyses to uncertainty in relationships, and whether taxonomic information can be a useful substitute for an explicit phylogenetic hypothesis where one does not exist. With this in mind, I collected data for 20 animal clades (vertebrate and invertebrate) that include fossil taxa, for which a recent cladogram and pre-cladistic taxonomies were available. I measured a series of phylogenetically explicit parameters (e.g. phylogenetic clustering of extinction [Fritz and Purvis' D], phylogenetic conservatism [Blomberg's K], inferred divergence times) and determined the degree to which they co-varied for taxonomic and cladistic trees. Results show that pairwise distances between taxa in taxonomies match those for formal solutions very well (R2 > 0.9 for all clades). With the exception of measures of phylogenetic clustering of extinction — which are sensitive to differences in tree topology — when taxonomic information is used, results of analyses are strongly correlated with those obtained using the formal solution and are therefore unlikely to be misleading relative to this standard. The particular analysis performed and the quality of the taxonomic information used must be carefully considered, but under some circumstances taxonomies are 'good enough' to be used in place of formal cladistic solutions.

D23SY24PS1387

STINGLESS BEE NEST EVOLUTION IS CHARACTERIZED BY MULTIPLE GAIN AND LOSS OF BROOD CELLS COHESIVENESS

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Stingless bees can construct comb or cluster nests. Cluster-brood building species construct interspaced clusters of discoid, vertically elongated brood cells with gaps in between cells. Conversely, the comb-brood building species make hexagonal, vertically oriented brood cells arranged in layers of juxtaposed cells showing higher cohesiveness. Cluster nest has been hypothesized as the basal state based on morphology and classical taxonomy. Nevertheless, that hypothesis has not been formally evaluated with modern phylogenetic and comparative methods. We estimated the ancestral nest state for each node in a large phylogenetic tree containing stingless bees of the New World (NW) and Old World (OW) in order to undercover the stingless bee nest evolution. Nucleotide sequences of five genes were retrieved from GenBank and concatenated. Substitution models were chosen with MrModelTest. We estimated the phylogenetic hypotheses with MrBayes and checked the convergence of parameters in Tracer/AWTY. We have confidently assigned the nest type for 101 out of 111 OTUs. We chose the MCMC option in BayesTraits for estimating the rate parameters. Analyses strongly support the cluster nest as the ancestral state of all stingless bees. The comb nest derived at least five times, once in the NW. The cluster nest would have re-evolved at least four times from comb nest ancestors exclusively in the Neotropics. The current hypothesis about the stingless bee nest evolution is in agreement with our ancestral estimations, although we formally provide the evolutionary steps. Therefore, increasing in cohesiveness of brood cells would be a derived feature that has independently evolved several times. The re-evolution of cluster nests would reflect a recurrent loss of cohesiveness between brood cells. The next step is to identify correlations between the age of ancestors and the paleovegetation in the hopes of shedding light into the selective forces shaping the convergence of the nest types.

Symposium

25. New Directions in Sex Role Research

21 August



Program

Wednesday 21 August

Session(s): 5, 6, 7, 8

Organisers: Tamás Székely and Michael Jennions

Invited speakers: Hanna Kokko and Larry J. Young

Description:

The evolution of sex roles is a core area in evolutionary biology linking life history and sexual selection theory. Recent advances in theory, experimental tests and phylogenetic analyses have provided fresh insights into sex roles. The Symposium will focus on 3 aspects of these recent developments.

D21SY25IT10:30R7

SEX DIFFERENCES IN THE EVOLUTION AND NEURAL MECHANISMS OF PAIR BONDING IN MONOGAMOUS PRAIRIE VOLES

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Monogamous prairie voles (*Microtus ochragaster*) provide an excellent opportunity to investigate the neural mechanisms underlying pair bonding behavior, as well as the evolution of those mechanisms. Behavioral pharmacological studies have demonstrated that oxytocin, a neuropeptide involved in parturition, milk ejection during nursing, and the onset of maternal nurturing and mother-infant bonding, plays a critical role in the formation of the pair bond between the female to the male. By contrast, the related neuropeptide, vasopressin, a sexually dimorphic peptide involved in territorial aggression and scent marking, plays a critical role in pair bonding in male prairie voles. The neural distribution of oxytocin and vasopressin receptors in the brain is strikingly different between monogamous prairie vole and non-monogamous species, being concentrated in dopaminergic reward centers in the prairie vole brain. Thus neural mechanisms underlying pair bonding evolved through adaptation of systems involved in maternal nurturing and bonding in females, and through adaptation of systems involved in territorial behavior in males. I will discuss molecular mechanisms by which diversity in neuropeptide receptor expression patterns leads to diversity in pair bonding behaviors in a sex specific manner.

D21SY25IT11:18R7

OPERATIONAL SEX RATIOS, BATEMAN GRADIENTS, COMPETITION, CARE, AND THEN SOME NASTY INTERFERENCE FROM THE DEEP EVOLUTIONARY PAST

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Reviews of sexual selection often emphasize one of two concepts. Strong competition for mates is expected when the operational sex ratio (OSR) is biased towards the focal sex, or when the Bateman gradient is steep for the focal sex. Treatments rarely emphasize that not one but two conditions must be satisfied before one expects selection for a trait that enhances mate acquisition: mating multiply must be beneficial for fitness, and achieving this must be difficult without the trait. Because the former idea is captured by the Bateman gradient and the latter by the OSR, the two concepts are complementary, not competing or mutually exclusive. Combining their insights into a single metric can help understand sex roles, but we also need to know the origin of OSR biases. These biases are often related to the way parental duties are divided or shared. The evolution of care roles is an outcome of a mix of positive and negative feedbacks involving e.g. the population-wide adult sex ratio. Together with the fact that it is easier for a parent to provide further care if it is already associated with its young (e.g., a parent that must physically be present at birth is a more likely caregiver later too), this implies that care evolution can feature strong phylogenetic inertia and multiple stable states. This makes care evolution challenging to analyze, but also offers new avenues for understanding e.g. male-biased care in fishes.

D21SY25RT14:00R7

DISSECTING THE BATEMAN GRADIENT: WHAT IT REALLY TELLS US ABOUT SEX ROLES

Julie Collet¹, Rebecca Dean², Kirsty Worley⁴, David S Richardson⁴, Tommaso Pizzari³
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At the heart of the recent debate on sex roles lies the interpretation of the Bateman gradient, the slope of the regression of reproductive success (number of offspring, T) over mating success (number of mates, M). Typically, males are considered to have a steeper Bateman gradient, thus to undergo stronger sexual selection than females. Recent work challenges this paradigm by questioning the measurement and interpretation of Bateman gradients. In this study we combine an experimental approach with multivariate analyses to resolve the significance of male and female Bateman gradients in the red junglefowl, *Gallus gallus*. First, the male Bateman gradient is measured by deducing mating success from paternity of the offspring, without accounting for matings that fail to result in fertilisation. We demonstrated that inferring M from offspring parentage leads to a 50% overestimate of the male Bateman gradient compared to a regression using fine-grain mating behaviour. Second, while the male Bateman gradient is concerned with the causal relationship between M and T, we show that variation in other components of male reproductive success, namely female fecundity and paternity share, also cause the Bateman gradient to be overestimated of a further 69%. Finally, there is growing appreciation that female Bateman gradients can be steeper than originally thought. We show that females display a positive Bateman gradient, suggesting that they benefit by mating with multiple males. However, an experimental test shows no evidence that productivity increases with number of mates in females, suggesting that the female Bateman gradient emerges as spurious consequence of males preferentially mating with more fecund females. Our results demonstrate that the mechanisms underpinning Bateman gradients are more complex than currently appreciated, and that understanding the causal relationship of M and T and how it defines sex roles requires an integrated experimental approach.

D21SY25RT14:24R7

HOMAGE TO BATEMAN: SEX ROLES PREDICT SEX DIFFERENCES IN SEXUAL SELECTION

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Classic sex role theory predicts that sexual selection should be stronger in males in taxa showing conventional sex roles and stronger in females in role reversed mating systems. I present the results of a study that tested this very central prediction and assessed the utility of different measures of sexual selection. We estimated sexual selection in both sexes in four seed beetle species with divergent sex roles using a novel experimental design. We found that sexual selection was sizeable in females, and that the strength of sexual selection in females and males varied with mating system and species. Residual selection formed a substantial component of net selection in both species. Further, we compared variance-based measures of sexual selection differentials) in their ability to predict sexual dimorphism in reproductive behavior and morphology across species. Our results 1) highlight the importance of using assays that incorporate components of fitness manifested after mating, and 2) allow us to identify the generally most informative measure of the strength of sexual selection in comparisons across sexes and/or species.

D21SY25RT14:48R7

FEMALE FINERIES REVISITED – ARE THEY CORRELATED WITH OFFSPRING QUALITY?

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1. The evolution and signalling content of female ornamentation has remained an enduring challenge to evolutionary biologists. While females usually invest significant amounts of their resources, including carotenoids, in offspring, all the resources allocated to elaborate ornamentation reduce resources available for other purposes. This may in turn constrain female fitness leading to dishonest female signalling.

2. We review the literature for empirical studies on mutually ornamented species with conventional sex roles, by focusing on the association between female ornaments and quality of their offspring.

3. We find 43 papers where 33 (77 %) are bird-studies, 9 (21 %) are on fishes, and 1 (2 %) is a lizardstudy. Nine of these report negative, 14 non-existing, and 20 positive associations between female ornament and offspring quality. Eighteen of the bird studies (55 %) show a positive association between the two traits investigated, whereas 5 (15 %) of the studies report a negative association. The number of fish studies, although few, is skewed in the opposite direction with 2 (22 %) and 4 (44 %) studies supporting positive and negative association, respectively. A minority of studies on carotenoidsbased ornaments reports a positive association (4 of 18 studies, or 22 %) between the traits, which is low compared to studies on non-carotenoids-based ornaments (16 of 25 studies, or 64 %).

4. The above mentioned relative large number of studies with negative association, especially common in studies on fishes and in carotenoids-based-ornaments, challenges the generality of the direct selection hypothesis to account for female fineries. This is important since this hypothesis seems to have strong support in the recent literature on the topic. In the present paper, we also propose possible explanations for the observed differences between taxa and suggest directions and ideas for the future research on the evolution of female ornamentation.

D21SY25RT15:12R7

SHOULD MALES INVEST MORE BECAUSE THEY ARE FATHERS? OR ARE THEY FATHERS BECAUSE THEY HAVE INVESTED MORE? THE EFFECT OF MALE ALLOCATION IN PARENTAL AND REPRODUCTIVE EFFORTS ON PATERNITY

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Although it has long been argued that males should adjust their parental behavior in response to female promiscuity level, empirical data does not support a general pattern of males behaviorally decreasing their parental effort in response to decreases in certainty of paternity. Here, we propose an alternative explanation, considering that male parental investment may have, in fact, played a more decisive role modulating paternity level over evolutionary time. In this sense, we predict that males experiencing high costs associated to care have been under stronger selective pressures favoring the evolution of post-copulatory processes that bias paternity towards parental males, in comparison to males that invest little in parental care. Although several game theoretical models evaluated the trade-off between precopulatory competition for mates and sperm competition, the additional component of male parental care has been largely neglected. Therefore, focusing in species with male-only care, in which females usually mate multiply and sperm transference is required by males prior to oviposition, we ask how males should allocate energy between mating effort, sperm expenditure and parental care. We extend previous sperm competition models and report the results of new theory investigating specifically: (a) whether the evolutionarily stable sperm allocation pattern can also explain the positive correlation between paternity and paternal effort predicted by classical theory; and (b) how female choice affects male allocation patterns.

D21SY25RT15:45R7

PARENT-GAMETE PROXIMITY AND ITS CONSEQUENCES FOR THE EVOLUTION OF SEX ROLES

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Why do some female marine animals release their eggs into the water to be fertilised, while others retain them on, or inside, their bodies? Why do terrestrial plants disperse pollen but not ovules, and why do most terrestrial animals hold onto both sperm and eggs until they find a mate? Sex role research has largely overlooked the significance of where gametes are located when fertilisation occurs. Parent-gamete proximity during fertilisation is not only a basic difference between the sexes in many species; it is also essential to the evolution of parental care and most forms of mate choice. I explore the evolutionary logic behind gamete release and retention using mathematical models. I focus particularly on marine invertebrates, in which evolutionary changes in these behaviours are common. Along the way, I provide a new explanation for why egg retention, small body size, and large egg size are correlated in sessile marine invertebrates. I also explain why there are no species in which males retain their gametes while females release them.

D21SY25RT16:09R7

SEXUAL SELECTION, SEXUAL CONFLICT AND FUNCTIONAL MECHANISMS OF POLLEN-PISTIL INTERACTIONS

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Despite theoretical and empirical evidence, sexual selection in plants has remained controversial for the past 30 years. Few plant studies have considered sexual conflict, even though this development of sexual selection has flourished in recent years. In our study species, the hermaphroditic annual Collinsia heterophylla, our experiments suggest a sexual conflict over timing of stigma receptivity. Delaying timing of stigma receptivity is advantageous for the female function in terms of enhanced pollen competition leading to increased offspring quantity and quality. However, early fertilization would benefit the male function as competition with later arriving pollen is avoided. Interestingly, our recent results indicate that male and female conflict traits may be interrelated within an individual plant, possibly suggesting trade-offs in sex allocation or a co-dependence between genes causing these effects. In other species, studies on pollen-pistil signalling mechanisms have mainly focused on incompatibility reactions between or within species or pollen tube growth and guidance in the pistil, rather than on sexual conflict/sexual selection. We are currently aiming to understand more about functional mechanisms of pollen-pistil interactions related to sexual conflict by analysing the transcriptomes (RNA-seq) at different stages of pistil and pollen interactions.

D21SY25RT16:33R7

PATERNITY, SEX ROLES AND FISH: WHO CARES?

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Female parental care predominates in most taxa because sperm competition and paternity uncertainty hamper the evolution of male care. What if fathers can, however, increase their share of paternity by providing parental care? Here we show that this provides an important modification to Dawkins and Carlisle's (1976) cruel bind – the idea that whichever parent releases their gametes first can desert the other, who is left 'holding the baby'. In our model, post-mating protection of paternity provides an extra incentive for males to stay with their young after gamete release. If offspring survive better when guarded, this paternity protection is enough to kick-start the evolution of male-only parental care from a no-care scenario. Our results help to explain the evolution of parental sex roles in fish, where male-only care (the norm) is associated with external fertilization, whilst female-only care almost always evolves after an initial transition to internal fertilization.

D21SY25RT16:57R7

SOCIAL ROLE SPECIALISATION PROMOTES COOPERATION

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Understanding the evolution of cooperation is crucial for understanding the evolution of breeding systems. An assumption implicit in almost all cooperation research is that cooperative behaviour in one specific ecological task evolves independently of other such tasks; however, this is often unlikely to be true. We use an individual-based simulation to relax this assumption, using biparental care as a model of cooperative behaviour. We show that synergistic costs of investing in two distinct care tasks, or a negligible sex-based asymmetry in their costs, select for sex-based task specialisation and stabilise cooperation between parents. Cooperation persists in spite of intense sexual selection and sex-biased mortality. Remarkably, with increased levels of cooperation, population size is also increased. We therefore show that ignoring the multivariate tasks animals face in nature leads to restrictive predictions of the ecological and demographic conditions under which biparental care in particular, and cooperation in general, are maintained.

D21SY25RT17:45R7

CONTEXT-DEPENDENT PLASTICITY OF REPRODUCTION IN THE COMMON GOBY (POMATOSCHISTUS MICROPS)

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Reproduction in animals can be highly variable with characters like reproductive rates and reproductive success being affected by numerous abiotic and biotic factors. Different populations of one species usually experience different kinds of such external influences. Likewise, there are often temporal changes in the environment, e.g. in the course of the progressing breeding season. By differentially affecting reproduction in males and females this social and population ecological context can also determine the direction and strength of sexual selection. While most research on sexual selection thus far has ignored this background, this study is specifically addressing the interactions between reproduction, sexual selection and its natural context, using the common goby (Pomatoschistus microps) as a model species. This small marine fish has exclusive paternal care, a resource-based mating system and plastic sex roles. Additionally, it occurs throughout the Baltic Sea and its distribution thus naturally stretches over steep environmental gradients. In this study, we investigated how different seawater temperatures and adult sex ratios affect reproduction, the operational sex ratio and intra- and intersexual competition in a controlled laboratory experiment. We found that warmer water led to higher reproductive rates in both sexes. However, this increase was much higher in males. Other preliminary results show larger clutches and higher reproductive rates under female-biased conditions. This study thus helps to further our understanding of the context-dependency of reproduction and we will discuss how this ultimately affects sexual selection.

D21SY25RT18:09R7

FEMALE AGE DEPENDENT RESPONSES TO A MALE SIGNAL CAN ALTER OPPORTUNITIES FOR SEXUAL ANTAGONISTIC CO-EVOLUTION

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Evolutionary explanations for ageing are based on the observation that the strength of selection often wanes with age. While this is well established for natural selection, data on the age-specific expression and benefits of sexually selected traits are lacking. Here we present work to address this omission. We report the responses of *Drosophila melanogaster* females of different ages to receipt of an ejaculate protein, the sex-peptide (SP) transferred to females during mating. SP can mediate sexual conflict as it can cause fitness costs in females, while simultaneously benefitting the males that transfer it. Virgin and mated females of all ages showed significantly reduced receptivity to further mating in response to SP. However increases in egg laying in response to receipt of SP were observed only in young virgin females. Hence there was a narrow demographic window in which males could maximise the fitness benefits gained by transfer of SP. The pattern of female responses to SP reversed with increasing female age, hence altering the opportunity for selection. The data reveal a new example of demographic variation in the strength of selection, with convergence and conflicts of interest between males and ageing females occurring over different facets of responses to a sexually antagonistic trait.

D21SY25RT18:33R7

EXTENDING SEX ROLE RESEARCH TO HERMAPHRODITISM: POST-COPULATORY ALTERATION OF MALE AND FEMALE FUNCTIONS VIA SEMINAL FLUID IN A FRESHWATER SNAIL

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There are generally two sex roles that can be expressed separately (males and females) or simultaneously (hermaphrodites). Several fundamental differences in reproduction between these two sexual systems invoke interesting questions for generalization of sex role research. For instance, in contrast to separate sexes, a hermaphrodite theoretically has the unique possibility to alter its mating partner's male as well as female functions to its own benefit. Here, we present the first study of such mate influencing in the great pond snail Lymnaea stagnalis. Previous work has shown that proteins in the seminal fluid delay egg mass production in sperm recipients, something that also becomes apparent from multiple mating experiments. We now report that this seems to be beneficial for sperm donors, as delayed egg mass production leads to more investment per egg. In addition, we found that recentlyinseminated sperm donors transfer half the amount of sperm to mating partners, which is also caused by male accessory gland products. Crucially, we reveal that, as a consequence, these donors obtain less paternity success. This decrease, which reduces the male function of recipients, would be beneficial for donors if recipients invest more in their female reproductive output in response. In other words, they seem to invest less in their ejaculate and more in their eggs. These two functions of seminal fluid in a hermaphrodite suggest their unique post-copulatory opportunities, in contrast to gonochorists. It would be interesting for future research to test if these post-copulatory effects of seminal fluid proteins alter evolutionary trajectories under various sexual selection scenarios.

D21SY25RT18:57R7

QUANTIFYING PRE- AND POST-COPULATORY EPISODES OF SEXUAL SELECTION IN A SIMULTANEOUS HERMAPHRODITE

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Classical sex roles are often assumed to apply to all sexually reproducing organisms with males being typically more eager to mate whereas females being more choosy. For simultaneous hermaphrodites this implies that mating should be more beneficial for the male than for the female sex function, but empirical tests are scarce. Specifically, a high mating activity (mating success) is expected to lead to the successful transfer of numerous sperm cells (insemination success) and thus to the fertilization of numerous eggs (fertilization success), jointly leading to a high male reproductive success. These consecutive episodes of sexual selection are often challenging to quantify separately, especially given the generally cryptic nature of post-copulatory processes, usually occurring inside the sperm recipient. A recently established transgenic line in the transparent flatworm *Macrostomum lignano* expresses green fluorescent protein (GFP) in all cell types, including the spermatozoa, which enables us to observe the mating interactions, the number of sperm received from a GFP-expressing mating partner and the resulting offspring produced (as the GFP locus is a dominant marker). We created 72 groups of 5 individuals, including one focal GFP-expressing individual, and assessed the mating success, the insemination success and the male and female reproductive success of these focals, of which we also measured a suite of morphological traits (including testis and ovary size, as well as the morphology of the male copulatory organ and the sperm cells). We aim to quantify (1) how mating success affects male and female reproductive success, (2) how mating success, insemination success and fertilization success contribute to male reproductive success, and (3) how these episodes of selection are affected by specific morphological traits. To our knowledge, this is the first study that quantifies all of the above mentioned episodes of sexual selection in a same individual.

POSTERS

D21SY25PS0074

SEX RATIOS FAVOUR AN EVOLUTIONARY TRANSITION IN SOCIAL BEHAVIOUR AND LIFE-HISTORY

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The evolution of eusociality, where some individuals refrain from reproduction to help others to reproduce, remains one of the biggest challenges for evolutionary theory. From a Darwinian perspective individuals are expected to maximize their own reproduction rather than help others reproduce. Different theoretical frameworks have been developed to explain why individuals would be selected to have reproductive altruism. In particular, kin selection theory has made salient the relevance of sex determination systems and sex ratios in the evolution of altruism. However, beyond changes in social behaviour, the transition between solitary and eusocial life involves also changes in life-history traits that have not been explain theoretically. Here we show that the coevolution of sex ratios and helping behaviour causes not only the evolution of eusociality, but also a transition between bivoltine and a univoltine life history. Our model shows that the haplodiploidy genetic systems can favour the evolution of eusociality, however this depends on the type of life history under which selection takes place. This is the first theoretical explanation for a life history transition during the evolution of eusociality. It points out to the importance of life-history conditions on the evolution of sex ratios and social behaviour.

D21SY25PS0106

NO EVIDENCE FOR SIZE-ASSORTATIVE MATING IN THE WILD DESPITE MUTUAL MATE CHOICE IN TWO SPECIES OF SEX-ROLE-REVERSED PIPEFISH

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Size-assortative mating is expected to be common in species with mutual preferences for body size. In this study, we investigated whether there is direct evidence for assortative mating in two species of pipefishes, *Syngnathus floridae* and *S. typhle*, that share the characteristics of male pregnancy, sex-role reversal and a polygynandrous mating system. We take advantage of microsatellite-based 'genetic-capture' techniques to match wild caught females with female genotypes reconstructed from broods of pregnant males and use these data to explore patterns of size-assortative mating in these species. We also developed a simulation model to explore the conditions under which preferences for body size can lead to size-assortative mating. Contrary to expectations, we were unable to find any evidence of size-assortative mating in either species. Results from simulations demonstrate that strong size-assortative mating preferences are unlikely to explain the observed patterns of mating in the studied populations. Our study suggests that individual mating preferences, as ascertained by laboratory-based mating trials, can be decoupled from realized patterns of mating in nature.

D21SY25PS0198

THE COMPOSITION OF THE SPERM MEMBRANE ACROSS MAMMALS IS AFFECTED BY MASS-SPECIFC METABOLIC RATE AND THE LEVEL OF SPERM COMPETITION

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Cellular membranes are key structures affecting cell function. Their characteristics and performance are strongly determined by the fatty-acid composition of the membrane phospholipids. Since higher levels of polyunsaturated fatty acids (PUFAs) are coupled with more metabolically active membranes, the "membrane pacemaker theory of metabolism" proposes that a high content in membrane PUFAs is positively correlated with high mass-specific metabolic rate (MSMR). This theory holds in many somatic cell types investigated so far. Spermatozoa are highly specialized and differentiated cells that undergo a long series of processes in the male and female reproductive tracts until they reach the site of fertilization. During this transit, they are prone to damage such as lipid peroxidation. We hypothesized that, in order to avoid such damage, and the ensuing impairment of sperm function, sperm cells exhibit a negative relationship between PUFA content and MSMR, since PUFAs are prone to lipid peroxidation, and more lipid peroxidation is likely to occur as metabolism increases. Given that high levels of sperm competition can lead to increased ATP production to fuel higher sperm velocities, we also hypothesized that higher levels of sperm competition would lead to a reduction in the proportion of sperm PUFAs. We performed two comparative studies on mammals (one literature-based, the other being experimental and comparing 4 *Mus* species that differ in their levels of sperm competition). We found that high MSMR and high levels of sperm competition both promote a decrease in the proportion of PUFAs that are more prone to lipid peroxidation. Our study indicates that the fatty-acid composition of membranes in sperm cells differs from that found in all other cell types, due to unique processes affecting the life of spermatozoa.

D21SY25PS0305

UNDERSTANDING PROMISCUITY: WHEN IS SEEKING ADDITIONAL MATES BETTER THAN GUARDING AN ALREADY FOUND ONE?

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Male monogamy is expected when a male spends his entire time budget guarding one female rather than acquiring multiple mates. The consensus from theoretical work is that mate-guarding intensifies with an increasing male bias in the adult sex ratio (ASR). Given that both male and female biased ASRs are observed, why is it that promiscuity is more common than male monogamy? We address this question with a model that allows males to combine paternity protection from guarding with searching for additional females to maximize paternity, i.e. temporally limited guarding. In contrast to existing models we combine pre- and postcopulatory mate guarding in a single model. Our model confirms that the ASR is an important predictor for mate guarding duration. Guarding durations increase with the number of male competitors and decrease with a decrease in female availability. However, a male biased ASR alone does not select for male monogamy. We identify conditions for promiscuity, temporally limited guarding and monogamy. These confirm our suspicions that frequency-dependent payoffs explain the relative infrequent occurrence of male monogamy.

D21SY25PS0368

SEXUAL SELECTION OF CHOOSINESS UNDER DIRECT BENEFITS

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Most theoretical research in sexual selection have studied the evolution of mate choice by indirect selection (*i.e.* when selection is only present on ornament and/or quality, but not directly on genes responsible for mate choice). However, empirical studies have not brought strong support to indirect selection. A less controversial finding is that choice is related to direct benefits and costs that exert a strong influence on the evolution of mate choice. We present an analytical model in which unilateral (female or male) choosiness evolves only according to such benefits and costs, *i.e.* only by direct sexual selection. We show that this simple model is sufficient to predict the evolution of all possible levels of choosiness when only four parameters are considered: the encounter rate, the lifetime and the length of unavailability after mating for males and females. This is because these parameters influence the trade-off between direct benefits in terms of the quality of mates and costs in terms of the quantity of mates. We further identify the sensitivity of the relative searching time (RST, *i.e.* the proportion of lifetime devoted to searching for mates) as a key variable allowing to predict the qualitative effect of any life history trait on the evolution of choosiness. Contrary to other predictors identified by previous models, the sensitivity of the RST encompasses the effect of all life history traits and constitutes an empirically accessible metric. The RST should allow a better understanding of the links between life history and mate choice in the future and thus provide new insights on the evolution of sex roles.

SEXUAL SELECTION CAN REDUCE MUTATION LOAD IN DROSOPHILA MELANOGASTER

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The 'cost of males' reduces the fitness of sexuals relative to asexuals. Sex is common, implying that the existence of males may generate indirect fitness benefits. One proposal is that deleterious alleles may be subject to stronger selection in males than females, due to sexual selection. This would eliminate harmful mutations at the expense of males and reduce the mutation load of sexual females. We performed competitive fitness assays to estimate selection on male and female fruit flies bearing chromosomes that accumulated spontaneous mutations. We found that selection on males was 75% stronger than on females, suggesting that sexual selection can reduce mutation load. We then asked whether the presence of parasites can enhance this effect. To examine the interaction of mutations and parasites, we assessed the fitness of mutant and non-mutant males and females that were inoculated with either sterile media or an opportunistic insect pathogen. Infection seemed to cause selection to be stronger in males and weaker in females. This interaction could increase the benefits of sexual selection. However, the presence of males could also reduce female fitness if many alleles are subject to sexually-antagonistic selection. When sexually-antagonistic alleles are common, a large fraction of the genetic variance in reproductive fitness should be due to intermediate-frequency alleles rather than rare deleterious mutations. We compared the contribution of deleterious mutations to standing genetic variance in male and female reproductive fitness and juvenile viability. We found that the fraction of standing variance explained by mutation varied among traits, but there was little evidence for sexual antagonism. Our experiments suggest that sexual selection can have a net positive impact on population mean fitness, with the potential to offset the cost of males in sexual populations.

REASSESSMENT OF BATEMAN'S PRINCIPLES: INSIGHTS FROM A META-ANALYSIS

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Bateman's principles have become a touchstone of sexual selection research by providing a catchy but highly controversial metaphor for the origin of evolutionary sex roles. Based on Bateman's classic study on *Drosophila* and further theoretical work by Trivers, Arnold and others, males are usually considered to be under stronger sexual selection than females because they typically show (1) more variance in reproductive success (RS), (2) more variance in mating success (MS), and (3) a stronger relationship between reproductive success and mating success (i.e., a steeper Bateman gradient). However, recent empirical work questions the universality of these so-called Bateman's principles. We take a meta-analytic approach to quantitatively test the hypotheses that the sexes differ in the variance in RS, the variance in MS, and the slope of the Bateman gradient across a broad range of plant and animal taxa. Furthermore, we explore how the behavioural mating system, the reproductive strategy, the type of fertilization and other factors modify the sex-difference in the three Bateman parameters, thus offering a life-history oriented understanding of the observed between-species variation.

D21SY25PS0492

POST-MATING SEXUAL SELECTION AND MAINTENANCE OF COLOUR POLYMORPHISM IN AN APOSEMATIC SPECIES

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Predation is assumed to select for signal uniformity and conspicuousness in aposematic species, but colour polymorphism is surprisingly common. Sexual selection can maintain colour polymorphism, but only a few studies have examined its role in the maintenance of polymorphism in aposematic species. Previous studies in the aposematic Wood Tiger moth, Parasemia plantaginis, suggest that the conspicuous yellow male morph is better defended against predators, whereas females seem to prefer to mate with the white morph offering one potential explanation for the maintenance of colour polymorphism. Here we test the possible direct benefits that female could gain by mating with white males by examining whether white males provide females with bigger spermatophores and more sperm and whether white males recover faster after mating than yellow males. Spermatophores of either white or yellow males were collected from recently mated females. To measure male recovery rate, the amount of fertile sperm stored in males' reproductive tract was assessed in mated males over several days post-copulation. We found that white males transferred bigger spermatophores than yellow males, but this difference was found only in older males. No difference in the pattern of sperm recovery was observed between male morphs. Contrary to our prediction, we found slightly higher number of fertile sperm stored in the reproductive tract of yellow males compared to white males, regardless of the recovery period. One explanation is that white males invest more in sperm competition by producing more non-fertile sperm that may suppress female receptivity. This would result in bigger spermatophores but with reduced number of fertile sperm. White males might also transfer other compounds such as defensive chemicals into the spermatophore that may benefit females. Altogether our results suggest that fertilization benefits do not solely explain how colour polymorphism is maintained in P. plantaginis.

FEMALE MOTHS DO IT ALL: ADVERTISE, COMPETE AND CHOOSE

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The concept of a male sex pheromone as a sexual trait that is used by females to evaluate males as mates is widely accepted. By contrast, the idea of a female sex pheromone as a secondary sexual trait is fairly new and evidence is scarce. In order to serve as an honest signal, sex pheromones, as other secondary traits that advertise condition, should bear a significant cost. Female sex pheromone, as opposed to male sex pheromone, is typically released in minute amounts. Nevertheless, accumulating evidence demonstrates an imposed cost on female produced pheromone. Male moths, at the other side of the sexual selection equation, are generally sperm limited and as such are expected to gain from choosing a mate. We will discuss the option of mutual mate choice and intrasexual competition among both, males and females in the context of operational sex ratio in monandrous and polyandrous females.

THE EFFECT OF SEXUAL SELECTION ON SEXUAL SIZE DIMORPHISM IN MAMMALS

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Rensch's well-known allometric law seeks to explain sexual size dimorphism across large groups of animal species. Rensch's rule states that, when males are the larger sex, the disparity between male and female body mass becomes greater as body mass increases. In a large scale study of birds Dale et al. (2007) revealed mating system as an important predictive factor in explaining size allometry. This suggests that Rensch's rule is driven by a correlated evolutionary response in one sex to stronger size selection in the other sex. Here, we investigate the influence of mating system on size allometry and Rensch's rule across the mammalian radiation. Across all mammals we find evidence for Rensch's rule only in polygynous species. This pattern is not recapitulated uniformly within the mammalian orders suggesting varying taxonomic responses to size selection. Using recently developed methods for detecting varying rates of phenotypic evolution we were also able to reveal how the specific evolutionary trajectory of body mass evolution differs between male and female mammalian species. Surprisingly, across all mammals we found that the rate of evolution was higher in females compared to males. This might reflect the idea by Lindenfors (2002) that sexual and fecundity selection can act as antagonistic selection pressures on body size in female mammals. However, this pattern is also variable at inspection of lower taxonomic groups.

DEEP RNA SEQUENCING SUGGESTS A LACK OF GLOBAL DOSAGE COMPENSATION IN THREESPINE STICKLEBACKS

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The establishment of non-recombining regions is a critical early step in the evolution of sex chromosomes. To compensate for the resulting difference in the expression of X-linked genes the heterogametic sex will increase the expression of X-linked genes or the homogametic sex will shut down one of the X chromosomes. This process is known as dosage compensation. It has recently been suggested that incomplete or imperfect dosage compensation may have a stronger influence on sexbiased expression than previously thought. We use the threespine stickleback (*Gasterosteus aculeatus*) as a model system to study the role of dosage compensation in the evolution and maintenance of sexbiased gene expression. Threespine sticklebacks constitute a particular interesting system in this respect because its chromosome group 19 are nascent sex chromosomes: Whereas there is still recombination between sexes in the region spanning the first 3 million bases on the chromosome, recombination is significantly reduced in the region from 3-12 million bases. The last 6 million bases are mostly deleted from the male Y-chromosome. We collected five specimens of each sex from a benthic and a limnetic population in four Alaskan lakes, respectively, and utilized deep RNA sequencing of brain tissue to study gene and transcript isoform expression differences between the sexes. Preliminary results from expression of genes corresponding to the missing region of the X chromosome suggest no dosage compensation as the expression pattern is indicative of a copy number variation biased towards females. In other areas of the genome, there is more varied sex-biased expression.

D21SY25PS0766

THE ROLE OF MATING BEHAVIOUR AND REPRODUCTIVE PHYSIOLOGY IN MATING SYSTEM EVOLUTION AMONG DANCE FLIES

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What explains diversity among mating systems is complex and controversial. One important determinant of sex differences in the reproductive behaviour (sex roles) is mating competition, influenced by the relative number of males and females available to mate (the operational sex ratio, OSR). The OSR is the result of sex differences in life history and can be influenced by the adult sex ratio (ASR) and mating latency. However, theory suggests that individuals with short periods of mating latency will be selected to mate more frequently regardless of OSR, and a biased OSR will not always drive a competitive response. There are few empirical studies that assess the relative importance of these factors for predicting transitions in mating system. Dance flies have highly diverse mating systems; with females of closely related species showing strikingly varied levels of sexually selected ornamentation. Adult females in many dance fly species receive all their dietary protein from "nuptial gifts" provided by males during mating. It is thought that competition for male donations has caused sex-role reversal in several lineages. I will present data suggesting that the OSR covaries with sex roles imperfectly in dance flies, and that time away from mating swarms is not the sole driver of sexual differences. Our capture-mark-recapture data reveal sex differences in swarm attendance in several dance fly species and suggest that while females from ornamented species visited mating swarms more frequently than those from unornamented species, male swarm visitation did not differ across mating systems. Finally, I consider the combined effect of ASR, swarm visitation frequency, mating frequency and some key characteristics of ovarian physiology on the OSR and mating system in several dance fly taxa. These data suggest a complex and inconsistent role for reproductive characters on the evolution of sex roles.

D21SY25PS0821

EXPLORING A FEMALE SEXUAL SIGNAL

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It is now well recognized that exaggerated sexual signalling traits occur in females across many taxa. However, it remains empirically true that such traits are less common in females than in males. Their relative rarity in females is attributed to more restrictive conditions under which they can evolve: theoretically, female investment into signals of mate quality should be restricted due to trade offs with investment into fecundity. The conditions necessary for the evolution and maintenance of exaggerated female traits are not fully understood and their exact signalling function is not always clear. We investigated the chemical signalling role of a conspicuous, female-limited trait in a beetle showing female courtship and male mate choice. *Megabruchidius tonkineus* females court males by repeatedly presenting their abdomen, which bears two large, dark, pore-enriched patches that the male probes and antennates before deciding whether to copulate. We manipulated female mating status, age, and size, measured male mating preferences and compared female chemical profiles to determine whether this exaggerated female trait functions to signal female fecundity.

A LIKELY ROLE FOR ANTAGONISTIC SELECTION IN THE MAINTENANCE OF GENETIC VARIATION AT THE ARGININE VASOPRESSIN RECEPTORS 1A

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Vasopressin modulates a range of socio-reproductive behavior in mammals, largely through its interaction with expression of the arginine vasopressin receptor 1a (avpr1a). In microtine voles, the length of the microsatellite locus situated in the avpr1a promoter region regulates expression of this gene in certain regions of the brain and this has a concomitant effect upon a variety of male-associated behaviors. We quantified the effect of variation in the length of the microsatellite in the promoter regions of avpr1a upon reproductive success in semi-natural populations of bank voles (Myodes glareolus). After artificial breeding was used to create lines with contrasting microsatellite genotypes, we released animals into 0.25 ha enclosures in the field and measured survival and reproductive success over a breeding season. Our study revealed a sex-by-density interaction for avpr1a microsatellite length, such that males with longer- and females with shorter- avpr1a microsatellite alleles enjoyed greater reproductive success in low density populations; conversely, males with shorter- and females with longer- avpr1a alleles produced more offspring in high density populations. Both sexually antagonistic selection and density dependent selection are reasonable candidate mechanisms behind the maintenance of variation in microsatellite allelic diversity in avpr1a.

ALL EGGS ARE MADE EQUAL

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Maternal effects mediated by egg size may have profound effects on offspring fitness. Sex-biased resource allocation in birds gains increasing interest, but it is not known to what extent the egg sexual size dimorphism (SSD) is a wide-spread phenomenon in this group. To answer that question we performed meta-analysis of 33 published and 2 unpublished studies, which included information on egg SSD of 31 avian species. Many of those studies suggested adaptive explanation for the reported egg SSD, which helped us to formulate predictions for our analyses. In some species, egg SSD was suggested to promote future size differences between the adults. If that is the case, then across species, adult SSD should be a significant predictor of egg SSD. However, in other species, egg SSD was invoked as an adaptive means by which a female balances nestling mortality differences between sexes, therefore producing bigger eggs for the smaller sex. Based on these two hypotheses, we derived a general prediction that there should be a significant relationship between the magnitude of adult SSD and the magnitude of egg SSD irrespective of the direction of those differences. Our analyses found no support for either of those hypotheses. Across species, adult SSD does not predict egg SSD. More importantly, our meta-analysis revealed no heterogeneity, with the meta-analytic mean very close to 0. That is, the observed variation in effect sizes in our dataset was almost exclusively explained by sampling error and there was no difference in avian egg sizes between the sexes whatsoever. Although adult SSD is undoubtedly a prominent feature of avian species, we conclude that, in general, there is no evidence for egg SSD across bird species.

D21SY25PS0920

CONTEXT-DEPENDENT PLASTICITY OF SEX-ROLES

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Most research on sexual selection thus far has been carried out while ignoring the social and population ecological context. Here I address the question how sexual selection and its natural context interact in a small annual marine fish with exclusive paternal care, the common goby (*Pomatoschistus microps*). Specifically, field experiments were carried out to study interactions between operational sex ratio, reproductive rates, mate competition and nest availability and its impact on reproductive decisions. The results elucidate the seasonal plasticity of sex roles in populations with changing environmental contexts and highlight the importance of nest density. Insights in temporal dynamics of the mating system interacting with the natural context and its potential population level evolutionary consequences shall be discussed.

D21SY25PS0969

THE EVOLUTION OF SEXUAL SEGREGATION WITHIN INFLORESCENCES: THE ROLE OF RESOURCE COMPETITION BY FLOWERS

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Spatial segregation of sexual functions within inflorescences has been explained as a mechanism of avoiding self-fertilization reducing the negative effects of inbreeding. However, many species with sexual functions separated in different flowers also have self-incompatibility systems making difficult generalizations of this hypothesis. An alternative hypothesis for this pattern is that flowers within inflorescences compete by resources, and that the sequential development of flowers and architectural constraints lead to a gradient on resource availability. For instance, fruit size also follows a positional pattern within inflorescences supporting this resource gradient. This variation on resource availability produces distinct optima for sex allocation accordingly with the position of the flower within an inflorescence, potentially driving the evolution of sexual specialization on separated flowers. Using as a model the largest family of flowering plants, Asteraceae, we explored whether floral specialization in sex functions (either male or female unisexual flowers) within inflorescences was related with higher levels of floral aggregation and therefore with higher resource competition between different flowers within the inflorescence. We measured number of flowers and capitulum size to estimate flower density in 101 species with different levels of sexual specialization: hermaphroditic (only bisexual flowers), gynomonoecious (bisexual and female flowers) and monoecious species (female and male flowers), using herbarium specimens from Swedish Natural History Museum and Coimbra University Herbarium. Statistical differences were assessed with phylogenetic generalized least squared models. Flower number and flower density at inflorescence level was correlated with the specialization degree on floral gender, suggesting that the increase of floral competition might favor sex specialization of male and female functions on different flowers within inflorescences.

RAPID CHANGE IN THE GENETIC ARCHITECTURE OF SEX-SPECIFIC FITNESS IN LABORATORY POPULATIONS OF FRUITFLIES

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Phenotypic plasticity requires the breakdown of genetic correlations between the phenotypes expressed in different environments. The same applies to sexual dimorphism, a conceptually related phenomenon where individuals express different phenotypes in response to the genetic environment provided by their sex-determining factors. While sexual dimorphism is ubiquitous, evidence is accumulating that its evolution is often incomplete. Thus, many populations harbour sexually antagonistic variation, alleles that increase the fitness of one sex at the expense of fitness of the other sex. This type of variation is evidence for genetic correlations between males and females constraining the evolution of dimorphism. While sexual antagonism has been documented in a growing range of animal and plant species, we know little about how adaptive conflicts between the sexes are resolved. Here, I present data on a change in the genetic architecture of fitness in laboratory populations of Drosophila melanogaster that is consistent with a resolution of sexual antagonism. We compared two populations that both descend from the outbred laboratory stock LHm in which sexual antagonism has been previously described. The populations have been separated for about 180 generations and maintained under highly standardised rearing conditions. Using estimates of genetic variances and covariances for male and female fitness in the two populations, we can show that the antagonism present in the ancestral LHm population has disappeared in one of the descendant populations. Thus, male and female fitness are no longer genetically correlated, despite the presence of genetic variation for fitness in both sexes and levels of variation at neutral genetic markers that are similar to those in the other population. Using genomewide genotype data, we are currently investigating the genetic changes associated with this rapid change in the genetic architecture of fitness and resolution of sexual antagonism.

SEX ROLES IN A DEMOGRAPHIC CONTEXT: POPULATION CONSEQUENCES OF INDIVIDUAL FITNESS VARIANCE IN SEQUENTIAL HERMAPHRODITES

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When studying sex roles, we often focus on which is the most competitive or choosy sex and who is performing more parental care. In sequentially hermaphroditic fish the division of reproductive tasks is as fair as it can get, since each individual (with few exceptions) reproduce initially as one sex and then change into the other sex, from female to male in protogyny (PG) or vice versa in protandry (PA). In either case, the second sex is the larger sex and potentially more prone to be removed by size-selective fishing. Does the direction of sex change influence the overall demography of a population? Sex change has been mainly investigated from adaptive/ecological points of view, focusing on individual variations in fitness between the male and female phase of the life cycle. Moving from the individual to the population level, the overall variance in individual reproductive success (Vk) should influence the demographic trajectory of the population. To our knowledge, no studies have compared protandrous and protogynous animal species in a population genetic context. Indeed, PG and PA can be expected to lead to very different demographic scenarios. PG occurs when there is high potential for polygamy and results in strong social structures dominated by one large male. Since large males can monopolize multiple females in harems and small females tend to choose the larger males, Vk should be greater in PG than in PA; this is expected to reduce the effective population size (Ne), a key parameter in conservation biology. This trend is reinforced by the greater skew in sex-ratio typically observed in PG. Here we show that molecular and life-history-based estimates of Ne in several pairs of protogynous and protandrous fish lend support to the prediction that protogynous populations may have - based on comparable trait distributions - generally smaller effective size than protandrous ones, leading to valuable insights for conservation and management of marine resources.

THE GENETIC BASIS OF CRYPTIC FEMALE CHOICE IN CHINOOK SALMON (ONCORHYNCHUS TSHAWYTSCHA)

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Cryptic female choice, a post-copulatory version of sexual selection little studied in external fertilisers, enables females to favour sperm of one conspecific male over another. In Chinook salmon, we have previously shown that female ovarian fluid differentially affects the sperm velocity of males in a female-dependent fashion, and thus that females may exert cryptic control of male reproductive success. Here we investigate whether this apparent form of cryptic female choice is 1) based on Major Histocompatibility Complex (MHC) dependent sperm selection or 2) influenced by overall genetic relatedness. MHC dependent mate choice is thought to have two main roles: i) to promote offspring with MHC allele combinations that increase immunological competence and/or ii) as a mechanism for differentiating kin, either to avoid inbreeding, or to preserve local adaptations. To reveal whether MHC compatibility is a contributor to the female-dependent sperm performance, we conducted paired-male competitive fertilisation trials with males of different sperm velocity in the focal female's ovarian fluid. After assessing the fertilisation success of each male via microsatellite based parentage assignment, we sequenced the parental MHC class I α and class II β loci and examined general relatedness of the parents with 9 microsatellites and a 6000 SNP Chinook salmon array (Clarke, unpublished). We show that sperm velocity is a key determinant for fertilisation success in Chinook salmon and is positively correlated with relatedness between mating pairs when the microsatellite data is considered. While preliminary at this stage, parental MHC genotypic distance, measured by nucleotide difference, does not strongly predict sperm velocity or fertilisation outcomes, but ongoing tests of associations to MHC amino-acid divergence, allelic counts and genome wide relatedness via SNP data might reveal more subtle patterns in cryptic female choice.

CAN MUTATIONAL LOAD BE REDUCED THROUGH SELECTION ON MALES?

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According to theoretical predictions sexual selection will purge mutational load and increase nonsexual fitness if most mutations are deleterious to both nonsexual fitness and condition-dependent traits affecting mating success. To test this hypothesis we manipulated the genetic quality of *Drosophila subobscura* males by inducing mutations with ionizing radiation and observed the effect of the obtained heterozygous mutations on mating behavior. We used the progeny of both groups (manipulated and non-manipulated) to test if sexual selection is efficient to reduce induced mutational load, by measuring nonsexual fitness. Within the non-manipulated and manipulated treatments we formed two groups with different opportunity for sexual selection, with presence or absence of female choice. Females mated more frequently with non-manipulated males and non-manipulated males courted females faster. Fecundity differences were obtained only in manipulated treatment. Group with the presence of female choice exhibited higher fecundity than group in which sexual selection was experimentally eliminated. There was no overall difference in egg-to-adult viability between different sexual selection regimes in any of the group. Our findings hint to an important role of sexual selection in purging deleterious mutations.

D21SY25PS1139

THE LANDSCAPE OF SEXUAL SELECTION

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Rugged fitness landscapes are thought to promote speciation, as they provide multiple adaptive peaks for populations to occupy. However, they have been rarely characterized, especially for sexual selection. Sexually selected fitness landscapes play a critical role in divergence of mating traits that underpins sexual isolation. We characterized the fitness landscape generated by sexual selection through male competition in stickleback fish. We capitalized on the highly variable genotypic and phenotypic combinations generated in an F2 mapping population to fully characterize the fitness landscape. This approach also allows us to experimentally tease apart selection on multiple traits. We measured fitness through male competition in naturalized habitats, assessing ability of males to acquire territories and defend nests, both of which are essential for access to females. We also measured male mating success. The species we study mate at different densities, which are expected to affect the strength of sexual selection. Thus, we manipulated density to ask whether fitness landscapes at low and high density differ, altering the direction of selection or the trait combinations favored. We then compared these fitness landscapes to naturally occurring phenotypic distributions to assess how sexual selection would affect evolutionary divergence in natural populations, leading to reproductive isolation and speciation. We thus fill a critical gap in our understanding of speciation.

SEXUAL CONFLICT PREDICTS THE EVOLUTION OF SEXUAL DIMORPHISM IN A DIVERSE AVIAN FAMILY, THE PENDULINE TITS (REMIZIDAE)

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Sexual selection is a potent evolutionary force that can cause the rapid diversification of morphological and behavioural traits. The Penduline tits (Remizidae) are a small passerine family distributed throughout Eurasia, Africa and North America which employ a diverse array of reproductive strategies. The European species (*Remiz pendulinus*) is highly promiscuous, with both males and females having up to 6 sequential mates in a single season. The pursuit of additional mating opportunities results in uni-parental care due to male and female nest desertion and, surprisingly common bi-parental desertion (up to 30%). The considerable variation in parental care conflict across this family led us to produce the first molecular phylogeny of the Remizidae family (13 species) so that the relationship between sexual dimorphism evolution and sexual conflict over care are associated with sexually dimorphic plumage traits. The relationship between parental investment and sexual selection is a key piller of sexual selection theory but the empirical examples of this in such closely related species are rare. To ascertain the drivers of this relationship is a key objective within evolutionary biology.

THE ROLE OF EXTRA-PAIR MATING AND COOPERATIVE BEHAVIOURS FOR THE EVOLUTION OF AVIAN MATING SYSTEMS

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There is a range of bird mating systems and sex roles, from true genetic monogamy, via social monogamy with extra-pair mating, lekking, and through to group breeding. We explain this continuum from ecological factors using evolutionary simulation models with typical reproductive trade-offs. The key mechanism is that extra-pair mating may trigger male-male cooperation, because extra-pair males benefit from diverting their behavioural efforts towards the neighbourhood where they potentially have offspring rather than monopolizing it towards their domestic nest. For species with high mortality, extra-pair mating and cooperative behaviours dominate, and where extra-pair mating is very high the model's predicted mating system resembles group breeding. For long-lived species there are two clusters of viable mating systems: lekking is predicted if females can raise the brood alone, whereas genetic monogamy is predicted where single parenting is unable to raise viable offspring, as in many seabirds. The prospect of predictively relating ecological factors to emergent breeding systems may have transferable value also for other taxa.

PROMISCUITY AND EVOLUTION OF COOPERATIVE NEIGHBOURHOODS

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A striking but unexplained pattern in biology is the widespread promiscuity in socially monogamous species. Although females commonly solicit extra-pair copulations, the adaptive reason has been elusive. We use evolutionary modelling of breeding ecology to show that females benefit because extra-pair paternity incentivizes males to shift focus from a single brood towards the entire neighbourhood, as they are likely to have offspring there. Male-male cooperation towards public goods and dear enemy effects of reduced territorial aggression evolve from selfish interests, and lead to safer and more productive neighbourhoods. Using a set of theoretical simulation models, we predict sexspecific cooperative behaviours at breeding sites where males cooperate more towards public goods than females. The mechanism provides adaptive explanations for the common empirical observations that females solicit extra-pair copulations and that neighbours dominate as extra-pair sires. Derived from ecological mechanisms, these new perspectives point towards the evolution of sociality in birds, mammals, and primates, including humans.

THE ROLE OF HOST SEX IN PARASITE EVOLUTION

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The most extreme inter-individual differences within species are often those between sexes. In populations of sexual species, these intersexual differences often include strong dimorphism in parasite prevalence, disease symptoms and virulence. These effects of host sex have traditionally been attributed to sex-specific differences in host properties such as behavior, immune responses, hormone balances and resource allocation. However, these profound differences between males and females may act as alternative "environments" for infectious parasites and may result in parasite lineages that are differently adapted to each host sex. Here, I will present different conceptual scenarios in which host sex can affect parasite evolution, illustrated with my empirical studies of bacteria evolving naturally in *Daphnia* or experimentally in *Drosophila*. A better understanding of specific adaptation of parasites to the host sexes will help us to understand sexual dimorphism in disease responses. Moreover, it will allow us to investigate how parasitism could favor the evolution of sexual dimorphism by reducing the probability that parasites could evolve optimal fitness across both host sexes.

D21SY25PS1428

TRANSCRIPTOME CHARACTERIZATION BY SSH-SEQ AND RNA-SEQ OF SPECIFIC TISSUES INVOLVED IN SEXUAL MECHANISMS IN AN ENDEMIC IBERIAN CYPRINID FISH

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Genetic mechanisms of sex determination are unexpectedly diverse and change rapidly during evolution, special among fish, and the diversity of determination systems can include environmental, genetic or even both components. Although one would assume that such a basic aspect of development as sex determination would be highly stable in evolution, the opposite can be found. The main players involved in such processes can normally be identified by screening for sex reversal mutations, functional studies and gender-specific expression patterns. Making use of these specific expressions, together with the fact that gonadal fate decision is not final but has to be actively maintained throughout life, the study of gene expression during gonad phenotype maintenance could provide clues for the processes behind sexual determination mechanisms. Squalius pyrenaicus is a helpful model to study sexual determination and differentiation in an endanger Iberian Peninsula endemic species that lives in a highly variable environment, presenting a balanced sex ratio. Through transcriptome sequencing using two different types of libraries, one produced with the standard Illumina RNA-seq and another previously enriched by Suppressive Subtractive Hybridization (SSH) technique, an extensive characterization of adult female and male transcriptome of this species was preformed, using organs mainly involved in sexual processes (gonad and brain, but also liver in libraries enriched by SSH) in order to identify genes that could be potentially be involved in sexual landmarks. Since S. pyrenaicus is the maternal ancestor and a genetic material donor to the allopolyploid Squalius alburnoides complex, this characterization will provide information to assess the conservation of these mechanisms in a hybrid context that frequently brings ploidy and genomic shock, abruptly changing genetic cascades, compared to those found in parental species.

HIGHER AGGRESSION TOWARDS CLOSER RELATIVES BY SOLDIER LARVAE IN A POLYEMBRYONIC WASP

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In the polyembryonic wasp *Copidosoma floridanum* females commonly lay one male and one female egg in a lepidopteran host. Both sexes proliferate clonally within the growing host larvae. Distinct larval castes develop from each wasp egg: the majority being 'reproductives' plus some 'soldiers' which sacrifice reproduction and attack competitors. Maturing mixed sex broods are usually female biased, as expected when intra-brood mating is common. Pre-mating dispersal followed by outbreeding is expected to increase sexual conflict over brood sex ratios and result in greater soldier attack rates. Due to sexually asymmetric relatedness, intra-brood conflicts are expected to be resolved primarily via female soldier attack. We observed soldier behaviour in vitro to test whether lower intra-brood relatedness (manipulated by whether or not the father was from the maternal population) increased inter-sexual aggression by female as well as male soldiers. As found in prior studies, females were more aggressive than males but, contrary to expectations, soldiers of both sexes showed more aggression towards more closely related embryos. We speculate that lower intra-brood relatedness indicates maternal outbreeding and may suggest a rarity of mating opportunities for reproductives maturing from the current brood, which may thus enhance the value of opposite sex brood-mates.

Symposium

26. The Genomic Architecture of Adaptive Traits: do QTNS Generally Exist?

20 August



Program

Tuesday 20 August

Session(s): 2, 3, 4

Organisers: Christopher W. Wheat

Invited speakers: Matt Rockmann, Thomas Mitchell-Olds and William

Cresko

Description:

This symposium is focused upon presenting a balanced perspective upon what we currently know about the genomic architecture of adaptive traits. While one part of the evolutionary genetic community is declaring that the QTN paradigm is a fruitless pursuit, others are finding genes of very large effects on fitness in the wild. Trying to generalize from these disparate findings is the goal of this symposia.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26IT14:48R7

QTNS OF NO EFFECT

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The effect-size distribution of QTNs is at the heart of much debate in the evolutionary genetics community. An important complication is that effect sizes are not intrinsic properties of alleles. They depend on environment and genetic background in complicated ways. Using *C. elegans* as a model, we have found that populations harbor extensive genetic variation with no effect on phenotype under ordinary conditions but with life-or-death consequences in perturbed conditions. Alleles with no phenotypic effect at all may contribute to adaptive evolution when background or environment changes. Given that alleles of all effect sizes clearly contribute to evolution, our research program should shift to identifying the biological circumstances that favor adaptation by specific genetic architectures.

D20SY26IT16:33R7

GENOMIC ARCHITECTURE FACILITATES AND FRUSTRATES THE IDENTIFICATION OF ADAPTIVE MUTATIONS IN STICKLEBACK

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Is adaptive evolution defined by numerous small mutations, few large mutations, or some combination of both? This deceptively simple question has bedeviled geneticists for years. A simple model is the sequential fixation of adaptive mutations in a single population subject to directional selection. Adaptive evolution is in reality a complex and dynamic process. Populations are structured geographically, and mutations are often subject to gene flow as well as temporally and spatially variable selection. Under such a mutation-migration-selection schema a variety of mutation types can occur, such as single nucleotide changes, insertion-deletions, and large chromosomal rearrangements. Thus, both the genetic and genomic architecture of traits can evolve in a metapopulation context. A consequence is that the present effect sizes of adaptive alleles are the outcome of an evolutionary process, not a description of the entire path. More practically, the genomic localization of large effect loci may be enhanced at the expense of understanding the order of mutational changes. Only recently have we acquired the requisite genomic tools to fully tackle this problem in natural populations. I will discuss our recent findings of the genetic and genomic architecture of adaptive mutations in threespine stickleback, including recent discoveries of significant structural variation and its role in very recent (<60 year) and rapid parallel evolution. I will show how this genomic architecture can facilitate the hunt for adaptive loci using GWAS in natural polymorphic populations, but then pose significant challenges for the identification of the evolutionary history of the precise mutational changes. Lastly, I will argue that fully understanding the genetic basis of adaptation would require a complete description of the entire adaptive process, and the answer to 'what are the sizes of mutational changes' may often be 'all of the above'.

D20SY26IT18:33R7

SELECTION ON THE GENES THAT CONTROL COMPLEX TRAITS

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How does environmental heterogeneity influence growth, reproduction, and fitness in genetically variable plant populations? In *Boechera*, a wild relative of *Arabidopsis*, we used genome-wide markers to quantify selection coefficients at polymorphic loci throughout the life cycle, across multiple sites and years. We found strong natural selection that varied among environments, with stronger selection on reproduction than on survival components of fitness. Next, we cloned a QTL that controls defensive chemistry, damage by herbivores, and fitness in nature. This gene encodes the first enzyme in the glucosinolate biosynthetic pathway, causing variation in chemical defense and herbivore damage, with subsequent effects on fitness. These ecological effects are driven by functional changes following gene duplication, and by two selectively favored amino acid changes in the proteins that they encode. These changes cause a gain of novel enzyme function, modulated by allelic differences in catalytic rate and gene copy number, which control survival and reproduction in nature. Finally, to understand biochemical control of complex trait variation, we examined the relationship between pathway flux and protein polymorphism in the enzymes responsible for glucosinolate biosynthesis. We perturbed the enzymes in the glucosinolate pathway, and showed that flux control is focused in the first enzymatic step, and that flux control of these defensive phenotypes is robust across environmental treatments. Furthermore, signatures of selection showed that this enzyme is the only one in the pathway that shows evidence of selection. Our results support the hypothesis that natural selection preferentially acts on enzymes with high control over flux and phenotype.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26RT14:00R7

THE GENOMIC ARCHITECTURE OF MORPHOLOGICAL, LIFE HISTORY AND FITNESS TRAITS IN TWO LONG TERM WILD POPULATIONS OF GREAT TITS (*PARUS MAJOR*) - CAN WE FIND EVIDENCE FOR GENES OF MAJOR EFFECT?

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Currently there is much debate, yet relatively little empirical data, on the nature of genetic architecture of quantitative traits in wild populations. Where is additive genetic variation located in the genome? Is trait variation influenced by many genes of small effect distributed throughout the genome, or by a few genes of major effect? Do the same loci cause phenotypic variation in different populations? Great tits (*Parus major*) have been studied extensively in long-term studies across Europe, and are consequently considered an ecological 'model organism'. Recently, a large number of genomic resources have been developed for the great tit, including a genetic linkage map with 5,500 SNPs. In this study, we use three different marker-based approaches (partitioning of additive genetic variance across genomic regions, pedigree-based quantitative trait locus mapping and genome wide association scans) to investigate the genetic architecture of morphological, life history and fitness traits in two long-term study populations of great tit - one in The Netherlands, and the other in the United Kingdom. We demonstrate that for most quantitative traits there is little evidence of genes of major effect, and that the two populations share very similar genomic architectures.

D20SY26RT14:24R7

FINDING QTNS IN GENES AFFECTING CRANIOFACIAL MORPHOLOGY IN WILD HOUSE MICE

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The understanding of the evolution of complex phenotypes, as for example, biological shape, depends on the identification of the genetic variants responsible for between-individual variation, being this the variation over which natural selection would act. For that reason, we used a natural-occurring hybrid system with the aim of identify genetic variants involved in craniofacial bone morphology. Hybrid mice from the Bavarian contact zone between Mus musculus musculus and M.m.domesticus were used in this study. Skull and mandible shape were measured with 3D landmarks and analyzed using Geometric morphometrics. The shape coordinates were reduced to principal component (PC) scores, which were used as phenotypes for the genome wide association mapping. With the aim of detect genes affecting specific regions of the bones, which are not detected by the complex shape changes represented by principal components, 3D pairwise distances between landmarks were also included in the mapping. 14 genetic regions, containing 24 genes were associated with PC axis and 11 regions with 32 genes were associated with pairwise distances. Several of the genes identified are known to be involved in bone morphogenesis suggesting that the association mapping recovers credible candidate regions. One region was identified as having general (association with PC1-16.5%) as well as specific effects in skull shape. We are now associating allelic differences in the candidate regions with shape differences across the hybrid zone to identify QTN candidates. Our results not only identified new genes possibly involved in skull morphogenesis, but also indicate that they are responsible for some of the morphological differences between two closely related species, and therefore are relevant for understanding the evolution of morphological divergence. Besides, we show that performing association studies with wild populations from a hybrid zone is a very promising tool for studying complex phenotypes.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26RT15:45R7

GENOMIC PATTERN OF SELECTION FOR ECOLOGICALLY IMPORTANT TRAIT - TRANSCRIPTOME ANALYSES OF THE BANK VOLE ARTIFICIAL SELECTION EXPERIMENT

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Although, a response to selection at the genomic level has been investigated for some traits, little is known about the genomics of adaptation in ecologically important traits in vertebrates. Here we compared transcriptomes of the bank voles (Myodes glareolus) selected for high swim-induced aerobic metabolism (A) with unselected controls (C). In 13th generation of selection, voles from 4 replicate A lines achieved 48% higher max rates of oxygen consumption than those from 4 C lines (means±SD; A:5.32±0.64, C:3.59±0.57 mlO2/min) and differed in several other morphophysiological and behavioral traits. Our aim was to characterize the differences in allele frequencies and the differences in expression level between A and C lines, and thus pinpoint genes contributing to phenotypic diversification. Using Illumina paired-end sequencing and *de novo* transcriptome assembling we constructed reference liver and heart transcriptomes. Sequences obtained for each line from pools of liver and heart RNA were mapped to the reference transcriptomes to detect SNPs and measure the expression level. On average 33.5 and 29.3 mln 100 bp reads per line were obtained for liver and heart respectively, which allowed reliable polymorphism detection in over 7 000 genes resulting in more than 80 000 SNPs. About 1 600 of these SNPs showed allele frequency ranges which did not overlap between A and C lines, about 15% more than obtained from simulations assuming the differentiation purely by drift. However, the average differences in frequencies were similar for simulations and the experiment. We also identified a modest number (10 in heart and 45 in liver) of genes that showed more than two-fold differences in expression between A and C lines. Our results show that the rapid phenotypic diversification is accompanied by only minor changes in allele frequencies or expression level making the presence of genes of large effect unlikely and indicating a highly polygenic basis of

the selected trait.

D20SY26RT16:09R7

INVERSIONS, SUPERGENES AND THE GENOMIC EVOLUTION OF LOCI OF LARGE EFFECT

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Supergenes are defined as clusters of genetic elements maintaining fitness-related traits maintained in high linkage disequilibrium in response to selection for specific combinations of those traits. Recent studies have uncovered that supergenes may be maintained by structural variation presumed to help avoid recombination and the deleterious effects of maladaptive trait combinations. Chromosomal inversions are well-known for their suppressing effect on recombination, and can lock together multiple beneficial gene variants controlling specific trait combinations. There are historical examples, as well as numerous new cases of structural variation associated with the maintenance of welldifferentiated morphotypes, ecotypes, or species in sympatry in diverse taxa. Chromosomal rearrangements are therefore important mechanisms of genomic architecture evolution, which merge the control of multiple beneficial traits under a simple inheritance. Thus they provide a way of building up loci of large phenotypic effect, often found to be associated with adaptive variation and radiations. But exactly how multiple beneficial traits become recruited within inversions at the population level is still unclear. In this talk, I will discuss the origins of rearrangement-associated adaptations from plant and animal taxa. I will highlight the role of the ecology of each individual trait and their different combinations, the role of the selection regimes underlying the fitness benefits of tight linkage, and the role of introgression. A continuum of genomic architectures underlies adaptive variation from stable polymorphisms within populations to ecotypes and to ecological speciation. Inversions and supergenes are therefore excellent genomic microcosms to improve our understanding of the process of adaptation and the ecology and tempo of the build-up of linkage disequilibrium between multiple traits.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26RT17:45R7

A SEXUAL ORNAMENT IN CHICKENS IS DETERMINED BY LARGE-EFFECT PLEIOTROPIC ALLELES AT *HAO1* AND *BMP2*, SELECTED DURING DOMESTICATION

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The genetic analysis of phenotypes and the identification of the causative underlying genes remains central to molecular and evolutionary biology. By utilizing the domestication process it is possible to exploit the large differences between domesticated animals and their wild counterparts to study both this and the mechanism of domestication itself. Domestication itself is characterized by strong directional selection, which can leave putative signatures of this selection present in the genome in the form of reduced heterozygosity (referred to as selective sweeps). We have generated multiple intercrosses and advanced intercrosses based on wild-derived and domestic chickens to fine-map genomic regions (or QTL) affecting a sexual ornament (one to less than 400kb in size). These regions have been over-laid with putative selective sweeps identified in domestic chickens (each approximately 40kb in length), and found to be significantly associated with them. By using expression QTL analysis, we show that two genes in the 400kb region, HAO1 and BMP2, are controlling multiple aspects of the domestication phenotype, from a sexual ornament to multiple life-history traits. Resequencing of these animals reveals four differentially-fixed polymorphisms between the parental lines exist in strongly conserved regions within the selective sweep present within this region, which are candidate causative QTN. This study demonstrates the potential for large-effect mutations in domestication, as well as the use of selective sweeps to identify putative QTN in such instances.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26RT18:09R7

THE GENETIC BASES OF BODY SHAPE ARCHITECTURE OF TWO SYMPATRIC MESOAMERICAN CICHLID SPECIES: A RAD-QTL APPROACH

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Determining the genetic bases of phenotypic adaptation and ultimately speciation is one of the major quests in evolutionary biology. Cichlid fish species flocks are a prime example of recent rapid radiations often associated with adaptive phenotypic divergence from a common ancestor in a short period of time. In these fishes divergence in eco-morphological traits, including body shape, colour, lips and jaws, are thought to underpin their diversification. The Midas cichlid species complex (Amphilophus) of Nicaragua provides one of the few known examples of sympatric speciation, where species have rapidly evolved different eco-morphologies in young crater lakes. In this study we use SNP marker genotypes generated using ddRAD sequencing and a combination of morphometric analyses to identify significant QTLs in ecologically divergent cichlid species with different body morphs: an elongated limnetic species (A. zaliosus) and a high-bodied benthic species (A. astorquii) inhabiting lake Apoyo. A total of 453 informative RAD markers were identified in 240 F2 hybrids. These markers were used to construct a genetic map for which 25 linkage groups were resolved. We found 27 segregating SNPs linked to QTLs. The QTL-linked genomic regions were annotated to identify genes that likely contribute to divergence in body shape in benthic and limnetic Midas cichlid sympatric species. These results underline the effectiveness of RAD-Seq as a tool for rapid and efficient generation of QTL-targeted and genome-wide marker data and a promising method for investigating the genomic bases of divergence in ecologically relevant traits.

POSTERS

THE GENETIC ARCHITECTURE OF A REPRODUCTIVE TRAIT IN A WILD BIRD POPULATION

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The timing of egg-laying (lay date) in wild birds is a key determinant of overall reproductive success, with some species timing their breeding events to coincide with peaks in the abundances of key prey species. Mistimed breeding events can result in trophic mismatches and subsequently lower chick survival and fledging success. A phenotypic shift to earlier laying has been observed in a population of great tits (*Parus major*) in response to the earlier emergence of their winter moth larvae prey, itself the result of climate warming. Lay date has been found to have a small but significant heritable component. This study combines several genetic analysis tools, including QTL mapping and Genome Wide Association Study (GWAS) techniques, to describe the genetic architecture of lay date. Variation in this trait appears to have a polygenic basis.

GENETIC EXCHANGE IN A MULTIGENOMIC FUNGUS; RHISOPHAGUS IRREGULARIS

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Rhizophagus irregularis is a model species of an arbuscular mycorrhizal fungi (AMF). The AMF forms symbiotic relationship with roots of land plants, improving plant growth and protecting plants against parasites. *R. irregularis* is a particularly important species of AMF because it colonizes roots of most of crop plants such as rice, potato and wheat. However, different isolates of this fungus can affect plant phenotype differently. Moreover, it recently has been shown that two isolates of AMF can exchange genetic material, a process that can alter both, plant and fungal phenotypes. *R. irregularis*, is a coenocytic organism, which means that many nuclei coexist and can move in the common cytoplasm. The genetic exchange between two AMF isolates occurs via vegetative hyphal fusion. However, unlike in most fungi AMF produces multinucleate spores and it has been shown that each isolate of R. irregularis carries genetically different nuclei, which are maintained in successive AMF generations. What is unknown is the fate of parental nuclei after the genetic exchange, how many parental nuclei are exchanged and whether the mix of nuclei is random. In addition the nuclei are exchange with the surrounding cytoplasm. This lead to a question whether mitochondria from both parental isolates are transmitted to the offspring and how can it influence the fungal and plant phenotypes.

26. The Genomic Architecture of Adaptative Traits: do QTNS Generally Exist?

D20SY26PS0496

ANNOTATED GENES AND NON-ANNOTATED GENOMES: USE OF MODEL ORGANISM GENE ANNOTATION INFORMATION FOR UNDERSTANDING THE MOLECULAR BASIS OF TRAITS OF ECOLOGICAL AND EVOLUTIONARY IMPORTANCE

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Recent advances in molecular technologies have opened up unprecedented opportunities for evolutionary biologists and ecologists to better understand the molecular basis of traits of ecological and evolutionary importance in almost any organism. Nevertheless, reliable and systematic inference of functionally relevant information from these masses of data remains challenging. In my poster, I will highlight how the Gene Ontology (GO) database can be of use in meeting this challenge. The GO provides a largely species-neutral source of information on the molecular function, biological role and cellular location of tens of thousands of gene products. As it is designed to be species neutral, the GO is well suited for cross-species use i.e. functional annotation derived from model organisms can be transferred to inferred orthologs in newly sequenced species. In other words, the GO can provide gene annotation information for species with non-annotated genomes. I will highlight the both the strengths and the current weaknesses of using GO for enhancing the understanding of molecular function in ecologically relevant species and present some examples of its use for evolutionary contexts

GENOME-WIDE ANALYSIS OF THE GENETIC ARCHITECTURE OF FLOWERING TIME AND FITNESS IN WHEAT EXPERIMENTAL POPULATIONS

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Flowering time is a major adaptive trait, as it allows plants to synchronize their vegetative cycle with optimal environmental conditions and thus to maximize their fitness. This trait has been studied over 20 generations in different experimental wheat populations, relying on three gene pools: two selfing pools obtained by pyramid crosses of two sets of 16 parents. The third is an outcrossing pool obtained by random crossing of 61 parents by the use of male sterility. These three pools have been dispatched over 12 sites in France, and then cultivated year after year in isolation, without migration or human selection. Fast evolution, both over time and in space, was noticed for flowering time. Using both shifts in allelic, or association genetics, these adaptations were in part explained by polymorphisms at key genes controlling vernalisation or photoperiod sensitivity (Rhoné et al. 2008, 2010). We recently developed an extended study of the outcrossing population, genotyping about 400 SSD lines with a 9k SNPs array (Chao et al. 2010), and performing an extended phenotyping of flowering time, under contrasted environmental conditions (variations in vernalization and photoperiod). With this highly recombinant and highly diverse population we describe the distribution of effects over detected QTNs, the major QTNs corresponding to previously described candidate genes. The detected QTNs x environment interactions highlight the genetics of local adaptation. Comparing major genes handled by plant breeders, and results obtained on wheat experimental populations, we will show how the genetic architecture of flowering time and its strong interaction with environment can explain the genetic architecture of fitness traits in natural populations.

THE GENOMIC ARCHITECTURE OF ADAPTIVE DIFFERENTIATION IN SEED DORMANCY AND GERMINATION TIMING BETWEEN TWO NATURAL POPULATIONS OF ARABIDOPSIS THALIANA

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Timing of germination is expected to influence plant fitness strongly, because it determines not only the environmental conditions for the emerging seedling but also sets the context for all subsequent life stages. We studied the genomic architecture and adaptive significance of differentiation in seed dormancy and germination timing between two natural populations of the annual herb Arabidopsis thaliana located close to the geographic limits of the native range (northern Sweden and central Italy). With recombinant inbred lines (RILs) derived from a cross between the two focal populations, we mapped QTL for germination traits. First, seed dormancy was estimated for more than 400 RILs and the two parental lines using seeds that had matured in the greenhouse. Second, the timing of germination at the Swedish field site and seedling establishment at both field sites were documented for 220 RILs and the two parental lines using seeds matured at the respective field site and planted at the time of seed dispersal in spring. The Italian genotype produced seeds with a markedly stronger dormancy and germinated later at the Swedish field site than did the Swedish genotype. At both field sites, the local genotype outperformed the non-local genotype in terms of seedling establishment. In Italy, seed dormancy was positively correlated with RIL establishment success, while in Sweden this relationship was negative. Of the 11 QTL identified, one affected all three germination traits (dormancy, timing, establishment). This QTL had a huge effect size compared to the other detected QTL, and co-locates with the known dormancy gene DOG1. The results demonstrate that genetically based differences in seed dormancy are associated with differences in timing of germination and successful seedling establishment at the sites of the source populations, and thereby contribute to adaptive differentiation among natural populations of *A. thaliana*.

GENETIC BASIS OF LEAF TRICHOME DENSITY IN NATURAL POPULATIONS OF ARABIDOPSIS THALIANA

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Trichomes are hair-like epidermal structures on plant surfaces that may contribute to herbivore deterrence, UV protection, and tolerance to drought. In the annual plant Arabidopsis thaliana, leaf trichome density varies considerably among populations. We examined the genetic basis of differences in trichome density between two populations located close to the northern (Sweden) and southern (Italy) margin of the native range. We planted parental lines and 400 recombinant inbred lines (RILs) derived from a cross between the two populations at the sites of both source populations. Seedlings were planted at the time of natural seedling establishment in the autumn, and trichome density was scored on leaves before flowering in spring. We mapped QTL for trichome density using a linkage map based on 360 SNPs spaced at ca. 1 cM intervals. We asked how many QTL contribute to differences in trichome density and what is the magnitude of their effects? Do QTL for trichome density co-locate with candidate genes known to influence trichome density? Can QTL effects be linked to mutations in these genes? Preliminary analyses detected 11 QTL influencing trichome density. Of these, one QTL on chromosome 2 was consistently detected across years and at both study sites, and explained between 19% and 44% of the variation in RIL means. This QTL was located in a genomic region harbouring two genes known to influence trichome density: ETC2 and TCL1. Sequence differences between the two parental lines in these two candidate genes corresponded to four and one amino acid, respectively. We will present an analysis of the functional significance of the sequence differences detected and of their geographic distribution. In ongoing work, we explore the adaptive significance of trichome density by connecting variation in trichome density to variation in other putatively adaptive traits and to fitness in the field.

SINGLE GENE POLYMORPHISM ALTERS THE FORAGING ACTIVITY OF A PEST LEPIDOPTERA

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The extension of agriculture areas, needed to meet the food needs of a growing world population, subjects the environment to important anthropic pressures and rapid changes. However, some phytophagous insect species from the wild habitats have taken advantage of these abundant and nutritious cultivated hosts, where they establish and become pests. A representative of a major insect pest family, the Noctuidae, is Sesamia nonagrioides moth, well known for its economical losses on maize in African and Mediterranean countries. This species shows two populations distinct by their ecological preferences: one is a pest found in maize fields, while the other is exclusively found on wild herbaceous host-plants. The qualitative and quantitative differences between wild and cultivated resources should involve the expression and the selection of different foraging strategies. Because foraging (for) is a candidate gene and a major modulator of foraging strategies in a variety of invertebrate species, it could be involved in the adaptation of phytophagous insects to cultivated plants. Following an integrative approach, we compared the crop and the wild populations of *S. nonagrioides*. We found that they differ in their foraging activity by measuring their tendency to change of food patches. We identified two allelic variants of the *for* gene respectively associated with specific gene expression rates. One of the variant was harboured by the wild population and the other was predominant in the crop population. Activating the encoded cGMP-dependent PKG by a pharmacological compound increased the foraging activity of larvae, which suggests a causal link between *for* genotypes and the identified behavioural phenotypes. To understand if the evolution of the for gene would be a factor of the adaptation of S. nonagrioides to cultivated host-plant, analysis of neutral markers and *for* polymorphism over the geographic and ecological range of the species is in progress.

THE X FACTOR: THE GENOMIC DISTRIBUTION OF EQTNS THAT REGULATE SEXUAL DIMORPHISM

<u>Rebecca F Dean</u>¹, Michael Stocks¹, Björn Rogell², Urban Friberg¹ ¹Department of Evolutionary Biology, University of Uppsala, Sweden ²Monash University, Australia becky.dean@gmail.com

How can sex differences arise from a largely shared genome? Theory predicts sex chromosome linkage is crucial: by reducing the intersexual genetic correlation, sex chromosomes allow males and females to evolve separately and reach their phenotypic optima. However, studies on the role of sex chromosomes and sexual dimorphism find mixed results: whereas sex chromosomes tend to harbor sex biased genes, the association between sexually dimorphic phenotypes and sex chromosomes is more tentative. We approach this question from a different perspective by examining the role of the X chromosome in the regulation of sexually dimorphic gene expression. Using the Drosophila Genetic Reference Panel we perform a genome-wide study to find SNPs that associate with variation in sexually dimorphic gene expression. First, we find the X chromosome is a hotspot for SNPs that associate with variation in sexually dimorphic expression, particularly when SNPs are located between genes. Furthermore, we show the far reach of the X chromosome - trans-regulating SNPs that associate with variation in sexual dimorphism are more common on the X chromosome. Finally, we look in fine detail at the genomic regions with dense dimorphism-associated SNPs to see whether we can identify individual SNPs as general regulators of sexual dimorphism. Taken together, these results suggest the X chromosome is a master regulator of sexual dimorphism and give more general insights into the genomic basis of complex phenotypes.

THE GENETIC ARCHITECTURE OF SKIN PRICKLING AND BODY SHAPE IN HYBRIDIZING SCULPINS (COTTUS)

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European freshwater sculpins, Cottus rhenanus and Cottus perifretum, have formed hybrid lineages (invasive Cottus) in the Lower River Rhine. The hybrid Cottus particularly resembles one of its ancestral species, C. perifretum, in phenotypic features. This contrasts with the expectation of hybrid intermediacy, which is suggested by the fact that the invasive genome is thoroughly admixed. Among these traits, the scale-like skin prickling and body shape represent a common source of variation among a number of Cottus species. The Ectodysplasin (EDA) signaling pathway provides promising candidates to investigate the genetic basis of prickling because it is known to affect the development of dermal bones and scales in fishes. We identified and mapped *Cottus* EDA signaling pathway components and performed quantitative trait loci (QTL) mapping for prickling and body shape in *Cottus*. A single highly significant QTL that affects prickling in all F2 mapping families was detected in an interval that contains the EDA receptor (Edar) with the maximum LOD score on *Cottus* linkage group 3 but none of the other EDA pathway genes. The same QTL region also shows effects on body shape. An investigation of gene structure from 6 individuals suggests that the genomic architecture of the Edar gene makes it a more likely contributor to evolutionary changes than other components in this pathway. An analysis of ancestral allele frequencies within EDA pathway gene regions in the invasive gene pool shows that Edar genomic ancestry is biased towards *C. perifretum* (85%-96%), which is correlated with the phenotypic similarity of invasive *Cottus* with that species. The Edar carrying QTL is currently the best candidate that strongly determines phenotypic variation of hybrid *Cottus* in the River Rhine. EDA signaling constitutes a key adaptive trait in stickleback and our results suggest that the same pathway may contribute to conspicuous phenotypic variation in *Cottus* as well.

Symposium

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

21 August



Program

Wednesday 21 August

Session(s): 5, 6, 7, 8

Organisers: Christophe Eizaguirre, Miguel Soares and Victor

Stiebens

Invited speakers: Simone Sommer, Jacob Höglund and Louis

Bernatchez

Description:

Conservation biology is one of the rare fields of biology where evolution has too often been neglected. The reason for this probably stems from the misconception that evolution does not act on an ecologically relevant time scale. Here, we aim to combine the fields of evolutionary (e.g. ecological genomics, host-parasite interactions) and conservation biology to demonstrate the necessity to consider evolutionary theories (e.g. effects of inbreeding, constraints on small populations) in conservation programs.

D21SY27IT10:30R6

WHY GENETICS, GENOMICS, BIOINFORMATICS AND EVOLUTIONARY THEORY MAY MATTER FOR CONSERVATION

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Conservation biology and evolution may at fist sight be a contradiction in terms. Evolutionary Biology is about how life on planet Earth has changed and Conservation Biology is concerned about to halt and stop some of the unwanted changes brought about by human impact. In this talk I will argue that to preserve biodiversity and halt the ongoing extinction of populations and species, a fuller understanding of the evolutionary processes brought about by human activities on natural populations is needed. It is now often acknowledged that a useful conservation strategy is to preserve any natural population's ability to adapt to future change. It is however, less clear how such a conservation program should be implemented. It has been claimed that in order to accomplish this task, conservation programs need to preserve genetic variation within the focal populations. It is however not clear what is meant by 'genetic variation'. I will discuss different approaches how genetic variation can be studied in a conservation context and how genomic data can be used to address issues of genetic variation relevant for conservation using examples from our own research.

D21SY27IT14:00R6

ECOLOGY, IMMUNOGENOMICS AND PATHOGEN RESISTANCE IN WILDLIFE POPULATIONS

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In order to protect functional biodiversity and ecosystem services we need healthy populations depending on healthy environments. Understanding the complex interactions that occur between human-induced environmental changes, pathogen pressure and transmission, the potential of host adaptive genetic diversity in host-pathogen co-evolutionary processes and disease resistance is a challenging task. It requires an interdisciplinary approach which should aim to integrate knowledge from evolutionary theory, behavioral ecology, conservation biology and conservation genomics. Next generation sequencing is revolutionizing conservation research and the new technologies are likely to replace traditional genotyping methods to a great extent in the near future. We are able to address questions in non-model organism whose answers seemed out of reach just a few years ago. In my talk, I will illustrate recent advances, challenges and pitfalls of current developments in evolutionary conservation. Specifically, I will address 1) The effects of behavioral strategies in shaping the adaptive potential and its fitness-relevance 2) The effects of human induced changes on pathogen pressure 3) The functional importance of adaptive variability of endangered species in host-pathogen co-evolutionary processes and pathogen resistance 4) The current challenges and pitfalls of next generation sequencing approaches in conservation genomics

D21SY27IT15:45R6

APPLIED EVOLUTIONARY GENOMICS IN FISH CONSERVATION: SOME SUCCESS STORIES AND CHALLENGES FOR THE FUTURE

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Conservation genetics has been defined as the application of genetics to preserve species as dynamic entities capable of coping with environmental change. Towards this end, molecular data can play two fundamental roles. The first one, inventorial, pertains to documenting patterns and has driven much of what we have accomplished until now. The second, mechanistic, refers to deciphering evolutionary processes underlying those patterns, is still in its infancy and this is where most of us put big hopes in the use of modern, high throughput genomics methods. Ultimately, we aim at finding causal relationships between genetic variation, phenotypes and the environment to predict future dynamics of selectively important variation and potential for adaptation to new conditions. In this presentation, I will illustrate some of the progress that we have made towards this end from our own research on fish conservation and management, and will comment on the main challenges that remains to be circumvented, no matter the power and resolution of the current genomics methods at hands.

D21SY27RT11:18R6

APPLIED CONSERVATION GENOMICS IN THE WOLVERINE (GULO GULO)

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Natural selection may rapidly lead to local adaptation in genetically structured populations of threatened and administrated species. Investigators trying to find specific genetic variation important for conservation of such populations in non-model organisms have previously been restricted to using a small number of anonymous genetic markers or variation in a few candidate genes. With the advent of high throughput sequencing and genotyping it has now, for the first time, become possible to identify adaptive genetic variation at a genome wide scale. We will present data from a whole genome sequencing project and large scale genetic variation profiling of a mammalian predator, the wolverine (Gulo gulo). Scandinavian wolverine populations are subjected to habitat fragmentation and high mortality from poaching due to conflicts with livestock herders. Genetic monitoring is already an important part of the national conservation programmes for this species in Sweden and Norway but the resolution and applications of genetic data have been hampered by a lack of genomic information. By assembling the complete wolverine genome and characterising genome wide genetic variation by resequencing of several different individuals, we now take this system to a new level. We describe how these novel genomic resources are utilised to investigate genomic signatures of population subdivision, reconstruct kin structure and to infer effects of inbreeding depression in small and partially isolated populations. We also discuss how the insights gained from this project will be applied to practical conservation efforts and monitoring of this highly charismatic species in the Scandinavian countries.

D21SY27RT11:42R6

DOES REINTRODUCTION AFFECT THE MATING SYSTEMS OF THREATENED SPECIES? THE IMPACT OF INBREEDING IN REINTRODUCED POPULATIONS OF THE HIHI (NOTIOMYSTIS CINCTA)

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An ever increasing number of animal and plant species are becoming rare or extinct in the wild. Reintroduction is one of the most commonly used tools for the management of these threatened species. However, this technique has a number of genetic, demographic and behavioural repercussions for which we have relatively limited understanding. In this study, of the endemic and endangered New Zealand hihi (*Notiomystis cincta*), our aim is to elucidate the impact of reintroduction on their mating system and reproductive success. We show the effects of inbreeding on individual reproductive tactics, extra-pair paternity and reproductive success and how it shapes the maintenance and evolution of these traits in wild small populations.

D21SY27RT14:48R6

DECLINING POLLINATOR POPULATIONS – DO VIRUSES HOLD THE SMOKING GUN?

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The pollinating insects – a diverse group comprising honeybees, bumblebees, hoverflies and many more – form a key group that is both crucial for guaranteeing our food security and for maintaining natural biodiversity. Many pollinator populations have been in decline over recent years and decades. Besides the obvious factors of habitat loss and change of land usage, infectious diseases and parasites have increasingly come into the limelight as potential causes of these declines. Rapidly evolving RNA viruses may be of particular importance, as they have been shown to be at the highest risk of causing emerging diseases. At the same time, such viruses can also be seen as a population genetic tool for studying the history of pollinator populations: quickly evolving viruses that are closely associated with their hosts allow host demographics to be reconstructed in the past. Here, I have used Acute Bee Paralysis Virus to study the pathogen transmission dynamics within bumblebee species communities consisting of rare and common species. Phylodynamic analyses reveal that this virus is commonly transmitted between species and that past epidemics may have contributed to range contractions. Rare and isolated species showed a high pathogen load whereas common species where relatively unaffected. This illustrates how non-specific pathogens may be able to shape species communities and how such coevolving pathogens may be used as monitoring tools to infer the history of species communities.

D21SY27RT15:12R6

DECOUPLING POPULATION SIZE, EFFECTIVE POPULATION SIZE AND PATHOGEN PREVALENCE IN A PERIPHERY-CORE SYSTEM IN A MIGRATORY BIRD, THE CORNCRAKE

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Inbreeding and reduced genetic diversity are known to affect the capacity of an individual to resist pathogen infection so that a negative relationship between pathogen prevalence and heterozygosity is expected. Thus, a high susceptibility to pathogens may contribute to shorten extinction time in populations with low genetic diversity. At the range scale, peripheral populations tend to be smaller and more isolated than core populations and are generally more prone to incur the costs of reduced genetic diversity. Elevated extinction risk in peripheral populations is of particular concern for species with unfavourable conservation status. However, the relationships between census size, effective size (genetic diversity) and parasite prevalence is not straightforward because gene flow from the core may maintain genetic diversity and the ability to resist pathogens at the range periphery, even in declining populations. The Corncrake Crex crex, a short live migratory bird distributed across the Palearctic has shown rapid declines in Western Europe over recent decades while Eastern populations remained large. We analysed the relationship between genetic diversity, gene flow and avian malaria prevalence across a large part of the species range. We found a very low level of population structure and no evidence of reduced diversity in peripheral populations which was likely caused by a high dispersal across the breeding range. Furthermore, contrary to expectations, the parasite prevalence was 10 times higher in core populations compared to peripheral ones, even in areas of high bird density. Our results clearly show that decouplings may occur in periphery-core systems between census and effective population size, and between genetic diversity and pathogen prevalence. Species dispersal characteristics, population densities and the ecology of pathogen community are likely to strongly affect these relationships precluding the quick and easy forecasting of a populations fate.

D21SY27RT16:33R6

A GENOME-WIDE ASSOCIATION STUDY (GWAS) OF AGE AT MATURITY IN ATLANTIC SALMON: IMPLICATIONS FOR CONSERVATION AND MANAGEMENT

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In wild Atlantic salmon (Salmo salar), there is variation in the age at maturity within and between populations, as individuals can spend anywhere between one and five years feeding at sea before returning, often to their natal river, to spawn. The larger, late-maturing 'multi-sea winter' (MSW) individuals that have spent multiple years at sea prior to maturation are highly sought after by anglers. This life-history strategy has also been shown to be important from a biodiversity perspective, thus creating a management conflict. Knowledge of the genetic basis of age at maturity would provide a foundation for developing effective management strategies for conservation of this important lifehistory trait. The Atlantic salmon population of the Teno River in northern Finland is arguably the most biodiverse salmon population in the world from a life history strategy perspective, with more than 100 different life history strategies (combinations of river years, years at sea and repeat spawning) being recorded. It also has immense socioeconomic importance, both due to fishing tourism (15,000 anglers visit this remote region annually) as well as local and indigenous fisheries, with up to 60,000 individuals (60% of the annual run of ascending salmon) caught annually. Alarmingly, the proportion of late-maturing MSW fish has been declining in recent decades. A long term scale archive combined with the availability of an Atlantic salmon 7K Illumina® iSelect SNP-array provided an opportunity to conduct a genome-wide association study (GWAS) to identify genomic regions associated with age at maturity. By implementing genome-wide relatedness information to account for unexpected population structure, we identified several genomic regions harbouring loci significantly associated with age at maturity. The implications of the findings for conducting GWAS in wild populations, as well as for the practical management and conservation of exploited salmon populations will be presented.

D21SY27RT16:57R6

MAINTAINING FITNESS AND DIVERSITY IN CONSERVATION PROGRAMMES

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Conservation programmes aim at maximising the probability of survival of the populations of interest. The goals in these programmes are to minimise the loss of genetic diversity, which allows populations to adapt to changes, and to reduce the increase in inbreeding. An optimal strategy to achieve these goals is using optimal contributions, that is, calculating the number of offspring every individual should contribute to the next generation to minimise global coancestry. Previous results on neutral scenarios had shown that molecular coancestry can maintain more diversity than genealogical coancestry if the number of markers available is large. However, using optimal contributions may allow deleterious mutations to be maintained in the population, compromising the long-term viability of the population. We show here the results of three strategies to avoid the maintenance of deleterious mutations: 1) combining optimal contributions with inbred matings, to expose and eliminate recessive deleterious mutations by natural selection, in a process known as purging, 2) using molecular coancestry calculated without markers at lower frequencies, as those could in principle be linked to a deleterious allele, and 3) using a measure of coancestry based on runs of homozygosity. Our results are strongly dependent on the genetic architecture and the mutational model assumed. It seems that once contributions are calculated, there is hardly any effect on performing inbred or outbred matings, and we have not observed any case where inbred matings lead to a later recovery in fitness. Using molecular coancestry in the management of the population maintains a larger genetic diversity but leads to a lower fitness than using genealogical coancestry. Using markers at intermediate frequencies maintains a larger fitness, but a much lower diversity. Lastly, using runs of homozygosity to measure coancestry seems to provide the best balance to maintain diversity without losing much fitness.

D21SY27RT17:45R6

MUSEUM DNA REVEALS A DRASTIC AND RECENT BOTTLENECK IN THE SEYCHELLES WARBLER

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Understanding the population history of endangered species is important for interpreting contemporary ecological processes, and of conservation high importance. We use microsatellite markers and museum specimens to examine 140 years of genetic diversity change in the Seychelles warbler (Acrocephalus sechellensis), a model system for evolutionary and conservation biology. In the 1960s this species was reduced to ca 30 individuals on a single island, but it is unknown whether it has ever been widespread and abundant. Across museum and contemporary samples, we found a 25% reduction in genetic diversity, which resulted in significant genetic structure. Using simulations we show that the warbler was bottlenecked from a large, outbred population. An ancestral effective population size of over 2,000 was reduced to less than 50 within the last century, which most likely occurred due to human disturbance. The approaches used form a powerful way of inferring population history, thus helping us to understand ecological processes and inform conservation.

D21SY27RT18:09R6

GENETIC CHANGES IN LIFE HISTORY TRAITS IN RESPONSE TO UNINTENTIONAL SELECTION IN A SUPPORTIVE BREEDING

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Supportive breeding is one of the last conservation strategies to avoid species from extinction when pressures exerted are difficult to solve. However, captive breeding may generate undesirable effects such as inbreeding depression, loss of genetic diversity or genetic adaptation to captivity that could be transferred to wild reinforced populations when a supplementation program is associated with the captive breeding. Several recommendations have been proposed to limit these deleterious effects but empirical assessments of such strategies remain scarce. Particularly, it might be difficult equalizing founders contribution in species with high reproductive skews where few individuals secure most of the copulations. We investigated potential genetic changes over generations in a supportive breeding of a lekking bird, the Houbara bustard. Using quantitative genetics based on a large pedigree dataset, we found that unintentional selection was occurring on life-history traits, particularly traits linked with reproduction leading to an increase of the breeding values. The main cause of this process might be the poor contribution of some breeders to the offspring. The situation seems difficult to avoid during the "growth phase" of the captive breeding consisting in quickly constituting a sizeable flock of adult breeders. Nevertheless, unintentional selection was decreasing over years, emphasizing the effort of managers to favour contribution of poor breeders in offspring recruited in the captive breeding. Besides, best captive-born breeders sired progeny that better survives once released in the wild, which would be expected under sexual selection. Our results shed light on very fast and strong genetic changes in a captive program that follows worldwide used recommendations. Decreasing the growth rate in the very early stages of the captive breeding should allow maximizing the contribution of poor breeders and reduce the risk of genetic changes.

D21SY27RT18:33R6

INCORPORATING INTRASPECIFIC VARIATION IN CONSERVATION PRIORITIZATION: A MULTI-TAXA APPROACH

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Human-induced land use changes are causing rapid habitat fragmentation. Species are therefore exceedingly limited in shifting their ranges in response to climate change, and likely need to adapt in situ to changing climate conditions. A prudent strategy to maintain the ability of populations to adapt is

to focus conservation efforts on areas where levels of standing intraspecific variation are high. By doing so, the potential for an evolutionary response to environmental change is maximized. We used spatially explicit ecological modeling approaches in conjunction with environmental variables to model species distributions and patterns of genetic and morphological variation in seven Ecuadorian amphibian, bird, and mammal species. We then used reserve selection software to prioritize areas for conservation based on evolutionary process (intraspecific genetic and morphological variation) or biodiversity patterns (species-level diversity). Reserves selected using species-level data showed little overlap with those based on genetic and morphological variation. Priority areas for intraspecific variation were mainly located along the slopes of the Andes, and were largely concordant among species, but were not well represented in existing reserves. Our results imply that in order to maximize representation of intraspecific variation in reserves, genetic and morphological variation should be included in conservation prioritization. To test the general applicability of our conservation prioritization framework, we are now applying this approach to target areas with varying levels of disturbance, different environmental gradients, and at small to large scales across four continents. Preliminary analyses indicate that the approach may be particularly useful at large scales, whereas other approaches should inform small-scale prioritization efforts.

D21SY27RT18:57R6

A RANGE-WIDE CONSERVATION GENETIC STUDY OF LITTLE PENGUINS (EUDYPTULA MINOR): AUGMENTING POPULATION GENETICS AT NEUTRAL LOCI WITH AN ADAPTIVE IMMUNE GENE

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Neutral genetic markers, i.e. genes not directly targeted by selection, are frequently used to quantify genetic diversity of populations and often provide the basis for management plans. However, patterns of variation and divergence in adaptive traits such as immune competence are not always correlated with variation in neutral markers. The validity of approaches using only neutral markers for development of conservation strategies is therefore questionable. The present study of conservation genetics and demography in E. minor aims to assess connectivity between penguin colonies along the Australian coast of New South Wales (NSW). A fine-scale analysis of genetic structure is being conducted to determine the extent of dispersal among these colonies and compare their genetic variability. In addition to using neutral genetic markers (microsatellites and mitochondrial DNA), we present the first study investigating non-neutral genetic diversity in penguins. Sequencing of an immune gene of the major histocompatibility complex (MHC) showed high allelic diversity at the functionally relevant peptide binding groove of the MHC molecule in penguins from Western Australia (WA). It is thus an ideal candidate gene to augment the population genetic study in NSW. This new genetic marker will be used to facilitate the choice of management strategies for E. minor and related species with a particular focus on resilience to pathogenic threats and immunogenetic population viability.

POSTERS

CONSISTENT PATTERNS OF RETURN CROSS-SPECIES APPLICATION OF SNP CHIPS

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Recent advances in technology facilitated development of large sets of genetic markers for many taxa, though most often model or domestic organisms. Cross-species application of genomic technologies may allow for rapid marker discovery in wild relatives of taxa with well developed resources. We investigated cross-species application of three commercially available SNP chips (the OvineSNP50, BovineSNP50, and EquineSNP50 BeadChips) as a function of divergence time between the domestic source species and wild target species. Across all three chips we observed a consistent linear decrease in call rate (~1.5% per million years), while retention of polymorphisms showed an exponential decay. These results will allow researchers to predict the expected amplification rate and polymorphism of cross-species application for their taxa of interest, as well as provide a resource for estimating divergence times.

HYBRIDIZATION IN A WARMER WORLD: COMBINING APPROACHES

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Origin and evolution of reproductive isolation are controversial, yet widely unknown. Reproductive isolation is the key characteristic of a biological species, and a common prediction is that genetic divergence between species is positively correlated with reproductive isolation. Knowledge of the importance of different isolating barriers during lineage divergence, and the intensity with which they prevent genetic exchange, are essential to understand the processes that result in the splitting of species, and consequently, in the hybridization. Numerous studies addressing species responses to climate change, mostly in animals, have illustrated that many species have altered their geographic ranges in response to rising global temperatures. Range shifts are leading to an increased sympatry between recently diverged species, likely increasing the potential for hybridization. Different approaches such as speciation genetics, large-scale genomic, bioinformatics, and predictive models for estimating patterns of species by the invasion of close relatives in future sympatric distributions favored by climate change. Understanding the processes related with the colonization by nonnative species provides essential information for biodiversity management.

INTERACTIVE EFFECTS OF INBREEDING IN A SPECIALIZED PLANT-HERBIVORE INTERACTION

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Inbreeding causes inbreeding depression in plant resistance against herbivores, as well as in several fitness-related traits in the herbivores. Furthermore, plant inbreeding may affect herbivore performance due to reduced herbivore resistance or plant nutritional quality. In many natural plant-herbivore systems, both of the interacting species are likely to experience inbreeding and yet, interactive effects of inbreeding of both a host plant and its herbivore have not been extensively studied. We studied the effects of experimental inbreeding of a perennial host plant, Vincetoxicum hirundinaria, and its specialist herbivore, the moth *Abrostola asclepiadis*, on plant resistance and herbivore performance in four populations. We were particularly interested in how inbreeding of both the host plant and the herbivore affect host and herbivore inbreeding depression in resistance and performance. Our results demonstrate that the expression of inbreeding depression in herbivore performance depends on whether the herbivore was grown on an inbred or on an outbred host plant and this effect varied among herbivore populations. Inbreeding depression in plants was significantly higher when they were consumed by outbred compared to inbred herbivores. Finally, the expression of inbreeding depression in the host plant in terms of resistance varied among plant and herbivore populations. These findings demonstrate that in plant-herbivore interactions inbreeding depression of one species can be altered depending on the inbreeding of the interacting species. Furthermore, our results suggest that when herbivores are inbred, herbivore-induced selection against self-fertilization in plants may diminish.

MAINTAINING IMMUNOCOMPETENCE UNDER INBREEDING: THE CASE OF A SELFING VERTEBRATE

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Mating between close relatives (inbreeding) reduces genetic diversity, making all individuals equally susceptible to environmental changes or diseases but, surprisingly, some populations seem to manage well and even thrive with low diversity. We have investigated how populations cope with low genetic diversity in a unique self-fertilising vertebrate (the mangrove killifish; Kryptolebias marmoratus). Our work shows how maintaining genetic diversity in some parts of the genome (i.e. the immune-related MHC genes) can be particularly important in terms of survival of the species. Hermaphrodite mangrove killifish have high rates of inbreeding and are genetically very homogeneous, but are also able to outcross, thereby introducing new genetic diversity. By comparing genetic diversity at neutral parts of the genome (microsatellites), immune-related genes (MHC) and parasites between wild and laboratory reared fish, we found that those individuals that had undergone more generations of self-fertilization had lower MHC and microsatellite diversity, and carried more parasites. But we also found evidence that the distribution of MHC alleles was not random, suggesting that even few divergent alleles could offer a selective advantage, potentially helping to fight a wider array of pathogens. Thus, even low levels of outcrossing could produce enough variability in immune-system genes to better resist parasites. This research has an important implication for conservation: immune-related genes, such as the MHC genes, need to be considered in captive breeding programs to minimize the negative consequences of inbreeding in endangered species.

CAN WE USE MOLECULAR TECHNIQUES TO MEASURE INBREEDING IN WILD POPULATIONS OF HIGHLY INBRED, BOTTLENECKED SPECIES?

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Understanding the effects of inbreeding depression is important for viability assessment and effective management of rare and endangered species, but accurately measuring inbreeding in such species remains challenging. Highly inbred species that have experienced severe bottleneck events typically lack the behavioural pedigree information, high levels of genetic diversity and large population sizes (and thus sample sizes) required to accurately estimate individual inbreeding. This raises the question of whether investing resources into research in this area is worthwhile for conservation managers and, if so, what the best approach might be. We modeled the accuracy of pedigrees constructed using varying numbers of simulated microsatellite markers for the little spotted kiwi (Apteryx owenii), a species that has experienced a recent, severe bottleneck and that exhibits extremely low genetic variation. We found that the number of microsatellite markers required to reconstruct precise pedigrees and provide accurate inbreeding coefficients will be cost prohibitive for the majority of conservation studies. Alternative approaches, such as heterozygosity fitness correlations, would also require a prohibitively large number of neutral markers. Pairwise relatedness measures show promise for estimating inbreeding in this species and those with similar histories, but genomic measures such as runs of homozygosity (ROH) currently provide the most effective alternative for measuring inbreeding. We acknowledge that genomic methods may also prove too costly currently for many conservation programmes and emphasise that conservation managers should weigh the costs and benefits of alternative strategies prior to investing in genetic measures as part of their rescue strategies.

CAN REPRODUCTIVE BARRIERS MAINTAIN DIFFERENTIATION IN FACE OF GLOBAL CHANGES? A CASE STUDY IN DROSOPHILA SUBOBSCURA

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Species with wide distributions may be highly differentiated across contrasting environments. While gene flow may help maintain similarities among populations, local adaptation may lead to their divergence, as well as promote reproductive isolation, further fostering evolutionary diversification. The interplay between these two processes is of major importance for Conservation, as it will determine if populations differentiate or become more similar in the long run, as a result of environmental changes such as those imposed by man. In Drosophila subobscura, populations are differentiated along a latitudinal gradient, but recent evidence indicates that northern populations are becoming more similar to southern ones. An important issue is how much populations from the extremes of the cline differ in mating preferences, as this might contribute to reduce genetic introgression when populations meet. To address this, we analyzed the evolutionary dynamics of reproductive isolation of D. subobscura populations derived from the extremes of the European cline, while adapting to a common, laboratorial environment. We show that mating performance increased during laboratory adaptation. In general northern populations had a better performance than southern ones, and this difference was sustained across generations. Moreover northern females preferred mates from their own populations while southern females preferred males from the north. The assortative mating of the northern populations was stable through time, while disassortative mating of the southern populations faded away during laboratory evolution. Overall this study suggests that reproductive barriers may slow down the genetic introgression due to migration to the north, an important finding in evolutionary and conservation terms.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

D21SY27PS0228

GENETIC DIVERSITY AND POPULATION STRUCTURE OF THE COMMON FROG (RANA TEMPORARIA) IN THE TRENTINO REGION (SOUTH-EASTERN ALPS)

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Amphibians are facing a dramatic decline worldwide, due to their high susceptibility to perturbations and global change. A decrease in genetic diversity can lead to loss of adaptability, and it is often associated with reduction in fitness. Amphibians seem to be particularly prone to such genetic processes, and a growing body of research shows that many amphibian populations are experiencing a reduction in genetic variation. In this study we investigated the levels of genetic diversity and population differentiation of the common frog (Rana temporaria) in the alpine region of Trentino. Rana temporaria is a widespread amphibian species in Europe and is not currently considered threatened; nevertheless, in some part of its range it has recently experienced localized declines. We analyzed polymorphism in a set of 12 SSR loci, in about 25 populations spread over the entire area of interest. Levels of genetic diversity were comparable to those found in other European populations, but a relatively high degree of heterogeneity among sites was recorded. We detected an irregular population structure, though a first general subdivision may be recognized between the populations belonging to the orographic right and left side of the Adige river. Some populations of the central part of the area showed lower levels of genetic variability, together with relatively strong differentiation. This subarea is characterized by a wide and anthropized valley run through by the Adige river: the suitable habitat for *Rana temporaria* is limited to isolated patches within an inhospitable landscape matrix. In addition, this area largely overlaps with the distribution of *Rana dalmatina* in the Trentino region. We discussed the effects of population density and potential competition (in terms of pond occupancy) on the recorded patterns of genetic diversity and population structure, highlighting the genetic peculiarity (and therefore conservation value) of some isolated populations.

MARK-RECAPTURE AND GENETIC ESTIMATES OF POPULATION SIZE IN A PROTECTED MOTH

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Accurate knowledge of both effective population size (*N*e) and census population size (*N*) is crucial for the efficient management of endangered species. Here we estimate both parameters for the protected moth *Graellsia isabellae*. We used both mark-release-recapture methods and microsatellites variation to obtain estimates of *N* and *N*e for two Iberian populations: Puebla (Eastern Spain) and Ordesa (Pyrenees). A different *Ne/N* ratio for both populations was the result of similar census population size estimates, and contrasting effective population size values obtained by molecular single-point and temporal approaches. Demographic analyses and comparison between long and short-term approaches revealed that the main factor causing disparity between *N*e estimates was a bottleneck, most likely caused by anthropogenic deforestation in Ordesa, although the species currently dwells in an optimal habitat. By contrast, the larger Ne showed by the population at Puebla confirmed its demographic stability. We discus the vulnerability of these two distinct populations of this emblematic insect, as the *Ne/N* estimates provided useful insights into the loss of genetic diversity and evolutionary potential. Therefore, we propose the re-categorisation of *Graellsia isabellae* in some sub-national and the national Spanish Red Lists.

CHARACTERIZATION OF MHC-CLASS II DRB DIVERSITY IN THE LESSER ANTEATER (TAMANDUA TETRADACTYLA)

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Studies of immune gene variation in the Major Histocompatibility Complex (MHC) provide a powerful tool to investigate patterns of adaptative genetic diversity shaped by natural selection. The use of MHC makers in population genetics has grown to a great extant in the past few years. However, studies with non-model species are still challenging due to the lack of a references and information of loci number. The lesser anteater is a medium sized mammal of the order Pilosa. The species is geographically widespread, found throughout South America in several types of habitats such as grasslands, savannas, forests and wetlands. Such flexibility on habitat use may lead to local adaptations reflected in the species' MHC gene diversity, accounting for different parasite pressures in the environment. To understand the contribution of natural selection to *Tamandua tetradactyla* populations we described the diversity of the MHC Class II DRB exon 2 gene. At first, Single Strand Conformation Polymorphism (SSCP) was used to screen diversity in order to develop the optimal primer pair for sequential amplification (i.e., the one that catches most diversity and shows less null alleles). DRB exon 2 was amplified in ten gDNA samples from individuals trapped at different geographical locations using three primer sets. PCR products were run twice on SSCP gels and sequenced. A fragment of 198 base pairs (66 aminoacids) was obtained, and no indels were found. After choosing the best primer set, a total of 22 alleles were detected on the aminoacid level (TateDRB01 to TateDRB22), and 26 on the nucleotide level. Between four and eight alleles were found per individual (average of 4.7), indicating the presence of at least two DRB loci. Secondly, 50 samples derived from four different biomes of the species occurrence are currently analysed through Next Generation Sequencing (NGS) on a 454 Junior platform. This will allow the analyses of the genetic structure of MHC DRB gene in the species.

THE "OMICS" OF INBREEDING DEPRESSION: A GENE AND PROTEIN EXPRESSION STUDY OF CONDITIONAL LETHALITY

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The study of inbreeding depression is relevant for agriculture, medical sciences, conservation and evolutionary biology. Although the population genetic principles of inbreeding depression are well known, the functional genomic basis remains poorly characterised. Recent studies have described changes in the metabolome and total gene and protein expression in inbred organisms. Functional studies enable us to explore mechanistic questions, e.g. what causes detrimental interactions between inbred genomes and environmental conditions. Since the complex and polygenic nature of general inbreeding depression makes this a difficult task, we focus on one aspect, conditional lethals in the fruitfly model system (Drosophila melanogaster). We present a detailed study of changes in the transcriptome and the proteome upon the triggering of a conditional inbreeding event, involving cold sensitivity and lethality. This study suggest that the inbreeding-related cold sensitivity in this line is caused by misregulation of normal mechanisms of cold tolerance. Therefore, study of the responsible QTL migth be informative about genetic variation in cold tolerance in outbred populations. In addition, we show that some of the changes in gene expression are part of a general signature of inbreeding depression.

NATIVE AND INVASIVE PREDATORS CAUSE VARYING ANTI-PREDATOR RESPONSES IN MARINE CRUSTANCEAN (IDOTEA BALTICA)

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Invasive species have negative impacts on ecological communities and they are a major worldwide problem. The North American white-fingered mud crab (*Rhithropanopeus harrisii*) is aggressive invasive species that has newly colonized also Northern Baltic Sea. Its predation pressure is high towards littoral fauna, e.g. crustacean grazer *Idotea balthica*. We studied the reported inability of *I. baltica* to avoid the crab and found that the isopod had strong anti-predator response towards native predatory fish (the European perch *Perca fluviatilis*) but with lethal outcomes, it did not recognize invasive *R. harrisii* as predator. The response to invasive and native predators was studied also taking into account the personality variation of isopods, as they have been shown before having behaviour traits fulfilling the criteria of personality. We finally discuss the impacts that invasive predators have on rapid evolution of anti-predator behaviour.

SPECIES DIVERSITY IN FRENCH TRADITIONAL ORGANIC BREAD DOUGHS

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Microorganisms result from billion years of evolution, but research on microbial communities took off recently with the rise of genomics technics. Those one are notably useful to link species diversity, phylogeny and ecosystems functioning. In food-processing environments, species diversity has been characterized in cheese or meat but less extensively in bread. Microbiome of bread doughs has been already described in Italia and Belgium but never in France. Here, we have analyzed the species diversity in bread doughs of five French bakers interested by maintaining biodiversity. The dynamics of species diversity were studied along the bread making process and from one year to another. The density of Lactic Acid Bacteria (LAB) and Yeast was measured using classical microbiology. Species were identified by gene sequencing (5' region of rDNA 26S and ActI for yeasts, and rDNA 16S for LAB). Biochemical characteristics of (sour)doughs and bread were also measured. Dominant yeast species varied between two bakeries (Candida humilis, Sacharomyces bulderi, S. exiguus or Torulaspora delbrueckii) and also from one year to another. In most cases, the same major yeast species was observed all along the bread making process. By contrast, LAB specific diversity was low with the same dominant species (Lactobacillus sanfranciscensis) found in most bakeries over the two years. The ratio of LAB over yeasts was relatively stable during the bread making process and from one year to another but differed between bakeries. Biochemical properties differed between bakeries and were related to the yeast and LAB frequencies. Previous studies from other European countries indicate that most bakers only maintain S. cerevisiae as Yeast in sourdoughs and the commonly reported LAB: yeasts ratio is 100:1. Taken together, our results suggest that French bakers sourdoughs host an interestingly diverse flora. The links between microbial communities and bread-making practices will be discussed.

SNAKE HEAD SHAPE MIMICRY: IMPLICATIONS TO THE CONSERVATION OF THE ENDANGERED MIMIC

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Most research on the adaptive significance of warning signals has focused on the colouration and patterns of prey animals. However, behaviour, odour and body shape can also have signal functions and thereby reduce predators' willingness to attack defended prey. For example, European vipers all have a distinctive triangular head shape and they are all venomous. Several non-venomous snakes are known to flatten their heads (head triangulation) when disturbed. Also many Lepidopteran larvae enhance their resemblance to tree vipers by concealing their heads and inflating their thorax or abdomen to express a false, sometimes triangular-shaped head. Even though anecdotal evidence of significance of snake head mimicry is dated back to the Henry Bates (1862), the role of body shape recognition is rarely experimentally investigated. Here we present data from field experiments and show that the triangular head shape can be recognized and avoided by predators. We also discuss the significance of this finding on population dynamics of snakes and its application their conservation. The smooth snake (Coronella *austriaca*) is non-venomous endangered species. By head triangulation it mimics vipers (*Vipera sp.*) which are not always protected by law. Because vipers are heavily killed by humans, it is possible that this asymmetric conservation program will be flawed because deceptive mimicry only works if the relative density and frequency of model species is higher than mimics. Based on the experimental evidence, we suggest that vipers should be protected at least in the locations where they co-exist with endangered mimic species.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

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CHARACTERIZING AND TRACKING MHC VARIATION OF CHYRTRIDIOMYCOSIS AFFECTED CHIRICAHUA LEOPARD FROGS OVER TIME IN NATURAL POPULATIONS

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The amphibian disease chytridiomycosis caused by the fungus Batrachochytrium dendrobatidis (Bd) has contributed to the decline and extirpation of numerous populations of Chiricahua leopard frogs (Lithobates chiricahuensis), an endangered species native to the Southwestern United States and Northern Mexico. Conservation efforts have focused on captive rearing of wild-caught egg masses and subsequent release following metamorphosis (called head-starting) to increase population sizes across the species range. The survival rate of released individuals is, however, heavily dependent on their ability to combat chytridiomycosis. In captivity, populations of other species of leopard frogs show differential survival to experimental Bd infections dependent on their genetic variation in the major histocompatability complex (MHC), a highly variable gene family in vertebrates known to be important in pathogen resistance. We used single locus 454 pyrosequencing to characterize MHC genetic variation in head-started Chiricahua leopard frogs that will be released and monitored for one year to determine survival rates and identify alleles that confer an advantage against Bd in a natural environment. This information will be used to identify candidate resistance alleles and add MHC-based marker assisted selection to captive breeding efforts, with the goal of increasing the success rate of head-starting efforts and mitigating the impact of Bd on natural populations of both Chiricahua leopard frogs as well as other amphibian species affected by chrytridiomycosis. This is a case study of how evolutionary genetic analyses of adaptive markers can assist conservation efforts in captive breeding.

LINKING FIRE ADAPTATIONS WITH ENVIRONMENTAL NICHES: THEORETICAL AND PHYLOGENETIC PROSPECTS

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In fire-prone environments, the evolution of plant life-histories is thought to be strongly influenced by fire regimes and environmental conditions. Fire management practices should therefore be informed by a better understanding of how fire related life-history traits have been shaped by environmental changes throughout evolutionary history. Perennial plant species in fire-prone environments have developed a wide range of adaptations to fire, which differ in the source of new recruits following fire events. Resprouting species can survive fires regenerating vegetatively from protected buds. In contrast, individuals of "obligate seeder" species are killed by fire: the population will only persist through seeds stored either in the soil or in the canopy (serotiny). To better explain such diversity of strategies, we used a optimization modeling approach (Tonnabel et al. 2012) to predict how fire regime and environmental conditions influence fire life history evolution (in particular serotiny). We then confront the predictions with data about relationships between life-history traits and environment collected for the genus Leucadendron (Proteaceae) in the South-African fynbos. We use comparative analyses approaches to test whether the evolution of particular fire-adaptations is linked to occurrence in specific environments (i.e. fire regimes, precipitation, temperature...) across a newly reconstructed phylogeny. This analysis reveals that fire adaptations arose several times during the evolutionary history of the Leucadendron. As predicted, we find that the evolution of serotinous obligate seeders is associated with the evolution of ecological niches where water limitation is weaker than for all other strategies. Such findings can help understand which species will probably be most vulnerable to climate change that is supposed to lead to dryer conditions in the South-African fynbos.

RED GROUSE (LAGOPUS LAGOPUS SCOTICUS) IN IRELAND AND SCOTLAND: ADAPTIVE GENETIC DIVERGENCE AND IMPLICATIONS FOR CONSERVATION

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Adaptive genetic variation is central to the survival of endangered populations, because it forms the evolutionary potential to respond to future challenges in a changing environment. Divergence in genetic variation among populations, however, has significant implications for conservation purposes. We here present a classical applied conservation biology question and base our answer on evolutionary concepts like selection, drift and adaptation: In Ireland, the Red grouse (Lagopus lagopus scoticus) is believed to be in risk of extinction, whereas it is rather abundant in Scotland and England. It has been proposed to restock the fragmented Irish populations with individuals from Scotland. We address the question whether Red grouse populations in Ireland and Scotland show significant divergence in adaptive genetic variation. If so, they should be recognized as separated evolutionary lineages for conservation purposes. Using 454-pyrosequencing we analysed immune genes of the MHC class II (Major histocompatibility complex), which are popular candidate genes to study adaptive genetic diversity due to their key role in many biological processes like pathogen resistance and mate choice. We compared the results with a study on neutral microsatellite variation to distinguish between genetic drift and selection. We found clear footprints of selection acting on the MHC genes, as well as variation in the number of gene copies, another classical feature of MHC evolution. The differentiation of adaptive genetic variation between Ireland and Scotland was strong with an Fst of 0.11, and significantly higher than differentiation based on neutral microsatellite variation with an Fst of 0.068. Thus, we conclude that these large differences in adaptive genetic variation are not due to drift and we advise against the introduction of Scottish Red grouse to supplement Irish populations. Instead we propose that efforts should be made to increase gene flow between Irish subpopulations.

MAJOR HISTOCOMPATIBILITY COMPLEX VARIATION AND ITS EFFECTS ON FITNESS IN THE ENDANGERED RED WOLF (CANIS RUFUS)

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The major histocompatibility complex (MHC) is a highly variable gene family which plays a critical role in cellular immune response. As such, measuring MHC diversity and its effects on fitness in wild populations is important because it may allow us to identify variation associated with population viability. The focus of our study was to estimate endangered wild red wolf (*Canis rufus*) MHC variation and assess its relationship to fitness. In total, we sequenced 350 wild red wolves at the dog leukocyte antigen class II genes DRB1, DQA1, and DQB. We related MHC variation at these loci to fitness measures including lifetime reproductive success and survivorship, which were obtained through a pedigree consisting of 23 years of detailed life history data. Additionally, we evaluated if particular MHC haplotypes or heterozygosity were associated with incidences of sarcoptic mange (*Sarcoptes scabiei*). These results will provide insight into how MHC diversity influences fitness and pathogen resistance in a wild population. Future work will be expanded to include Toll-like receptor genes, a family of genes involved in the innate immune system.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

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MHC DIVERSITY & PARASITE ASSOCIATIONS IN FINNISH WOLVES

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Maintaining adaptive genetic variation is one of the key factors in the conservation of small populations. Major histocompatibility complex (MHC) is a gene dense region where substantial numbers of immune genes are located. Lowered genetic variation in MHC loci can lead to lowered immunity against pathogens, which may increase the risk of extinction of the population. Humans have recently caused major bottlenecks in wolf populations around the species' distribution range. In Finland wolf population started to decline in the late 19th century and the population included only a few individuals in 1920s. During the last two decades the number of individuals first increased then decreased again and now it is around 130 individuals. We studied the amount of genetic variation and natural selection in MHC loci of Finnish wolf population compared to neighboring Russian population. We sequenced the highly variable second exon from three MHC class II loci; DLA-DRB1, DLA-DQA1 and DLA-DQB1 among 243 Finnish and 38 Russian wolves. We used 17 neutrally evolving microsatellite markers as reference loci. We also studied the association of MHC class II loci to Echinococcus granulosus and Trichinella spp. infections among the Finnish wolf individuals. Our results show that the Finnish and Russian wolves do not substantially differ in their MHC diversities. The allele frequencies in the Finnish population are more equal than expected; also Tajima's *D* and Fu & Li F^{**} tests give significantly positive values implying balancing selection. Historical positive selection is also evident in all three loci in relative amount of synonymous and non-synonymous polymorphisms. Temporal analysis show that inbreeding coefficient has increased in neutral microsatellite markers but decreased in MHC loci during the last 15 years. We found that MHC homozygosity increases the risk of Trichinella* spp. infection among Finnish wolves but a specific DLA-DRB1 allele is associated with decreased risk.

LIVING ON THE EDGE: HOW PHILOPATRY MAINTAINS ADAPTIVE POTENTIAL

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Without genetic variation, species cannot cope with changing environments and evolution does not proceed. In endangered species, adaptive potential may be eroded by decreased population sizes and processes that further reduce gene flow such as philopatry and local adaptations. Here, we focused on the philopatric and endangered loggerhead sea turtle (Caretta caretta) nesting in Cape Verde as a model system to investigate the link between adaptive potential and philopatry. We produced a dataset of three complementary genomic regions to investigate female philopatric behaviour (mitochondrial DNA), male-mediated gene flow (microsatellites) and adaptive potential (major histocompatibility complex (MHC). Results revealed genetically distinct nesting colonies, indicating remarkably small-scale philopatric behaviour of both males and females. Furthermore, these colonies also harboured local pools of MHC alleles, especially at the margins of the population's distribution, which are therefore important reserves of additional diversity for the population. Meanwhile, directional male-mediated gene flow from the margins of distribution sustains the adaptive potential for the entire rookery. We therefore present the first evidence for a positive association between philopatry and locally adapted genomic regions. Contrary to expectation, we propose that philopatry conserves a high adaptive potential at the margins of a distribution, while asymmetric gene flow maintains genetic connectivity with the rest of the population.

EFFECTS OF SEASONAL CONDITIONS ON MORTALITY AND FERTILITY IN ASIAN ELEPHANTS: IMPLICATIONS FOR CONSERVATION

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In highly seasonal environments, many species maximise offspring survival by reproducing at the time of year with peak resource availability. However, elephants do not have a single breeding season with females undergoing reproductive cycles throughout the year. Whether females concentrate most births on periods of maximum offspring survival is unknown, as are the proximate factors that could affect seasonal variation in birth rate. Long-term effects of birth season on patterns of mortality and fertility are also yet to be investigated. These topics are of particular importance in elephants, which are extremely long-lived and endangered, as understanding variation in mortality and reproductive success could contribute towards conservation of the species. We use individual-based longitudinal data for 2350 semi-captive logging elephants from Myanmar that occupy regions with a tropical monsoon climate, to investigate immediate and long term fertility and mortality responses to season. We supplement this long-term data with a 1 year subsample of 70 elephants for which monthly measurements of body weight, body condition and stress hormone (cortisol) levels were collected, to investigate the physiological correlates of seasonal conditions. Our results show significant variation in probability of birth by month, and an interaction between birth order and probability of birth, with higher seasonality of births in first-born individuals. There was no long-term effect of birth month on mortality, but females born in the monsoon season subsequently had earlier peak in fertility and earlier age at last reproduction in comparison to those born in the cool or dry seasons. The seasonal pattern of births did not coincide with high rainfall periods, but was rather associated with seasonal variation in workload and also matches individual level variation in cortisol in the subsample. This is a rare test of effects of early conditions on individual fitness in a non-model species.

LIFE HISTORY, SELECTION AND EFFECTIVE POPULATION SIZE SHAPING EVOLUTION DURING COLONIZATION – LESSONS FROM DROSOPHILA MELANOGASTER

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Natural selection is able to produce rapid adaptive responses to sustained environmental change under propitious conditions: intense selection, abundant genetic variation, and large population sizes. Consider a scenario where these conditions are not met: a population migrates to a new environment; it encounters qualitatively novel nutrition, possibly including periods in which it starves. How will its life history evolve in response to such a new environment? Furthermore, its effective population size may be reduced in the course of migration or as a result of an initial lack of adaptation to the new environment. How will effective population size affect its evolutionary response to this new environment? If this population survives and its descendants migrate back to their ancestral environment, how will this preceding period of adaptation to a new environment affect its initial life history upon return to ancestral conditions, and then its subsequent evolution? To tackle these issues requires populations of known differentiated histories, populations on which selection (both forward and reverse) is imposed with contrasting population sizes. This project involves laboratory populations of *Drosophila melanogaster* with controlled and replicated histories of selection. The study of their evolutionary dynamics after the imposition of new selection regimes, both at small and at large population sizes, will help resolve the interaction between genetic drift and natural selection, as a function of both previous and present selective and demographic histories

INBREEDING AFFECTS SOCIAL INTERACTIONS IN PARENTS

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Direct fitness consequences of inbreeding depression, i.e. the reduction of fitness related traits such as growth and survival of inbred individuals, are well documented. However, there has been much less work on the indirect fitness consequences of inbreeding in social systems in which individuals rely on social interactions with inbred individuals. In particular, there is now a need to understand the effects of inbreeding on social interactions among family members in species with elaborate parental care. We investigated the effects of inbreeding on social interactions of parents in the burying beetle, *Nicrophorus vespilloides*. This species breeds on small vertebrate carcasses and the parents provide elaborate parental care including regurgitating food to begging larvae. We conducted two experiments using two-by-two factorial designs in both, in which we first test for effects of inbreeding in male and female parents on interactions between caring parents and second we test for effects of inbreeding in female parents and their offspring on parent-offspring interactions. We find that inbreeding has direct effects on parental behaviour of inbred males, increasing the amount of care they provide, but that it has no direct effects on females. In addition, we find indirect effects of inbreeding in that females provide more care when males or offspring are inbred. Finally, we find evidence for an effect of an interaction between the inbreeding status of two individuals in that the apparent compensation of the female relies on the females own inbreeding status. We conclude that inbreeding influences the social dynamics of parents with important consequences for reproductive success, and propose that these indirect effects may extend to other social situations.

LOSS OF VARIATION AT NEUTRAL AND MHC LOCI IN PEARY CARIBOU OVER 100 YEARS: DRIFT IS MORE IMPORTANT THAN SELECTION

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Neutral markers are often used to estimate genetic variation in vertebrate populations, however, neutral loci may not reliably indicate adaptability because they are unrelated to fitness by definition and may lose variation more rapidly or more slowly than functional genes. We used threatened Peary caribou (Rangifer tarandus pearyi) historical (n \sim 30 c. 1905) and contemporary (n \sim 50 c. 2001) samples to compare loss of genetic diversity at 8 neutral microsatellite loci and one functional Mhc locus over time. Although the Mhc locus appeared to be under balancing selection, both types of loci appeared to lose similar amounts of variation indicating that drift was more important than selection or allele frequency distributions in this population. Allele frequency distributions and evidence of gene conversion may indicate that variation could recover more quickly at Mhc loci than microsatellite loci.

EVOLUTIONARY CONSERVATION OF LARGE BLUE BUTTERFLIES

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The large blue butterflies (genus *Maculinea/Phengaris*) are icons of invertebrate conservation. Their rarity is to a large extent due to their complex life cycle during which they exploit both specific host plants and specific host Myrmica ants, which must therefore co-occur. Even when both hosts are present in the same area, there is massive variation in the probability of adoption by and survival in different host ant nests. Over the last few decades we have learnt a great deal about some of the underlying causes of this variation, for example as a result of coevolutionary arms-races in chemical mimicry and the effects of variation in host ant social structure. Over the same period, there have been several attempts to reintroduce large blue butterflies to areas from which they have disappeared, with varying degrees of success. Here we examine to what extent the success of large blue butterfly reintroductions has been increased by a deeper understanding of their evolution, and the genetic and evolutionary consequences of reintroduction programs. We also examine how the success of future conservation efforts could be enhanced based on our current knowledge of the evolution and ecology of the large blue butterflies and their hosts.

COMPARISON OF ESTIMATORS OF EFFECTIVE POPULATION SIZE: VARIATION AND BIAS IN THE METHODS

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The effective population size (Ne) is a parameter of paramount importance in population genetics. Many estimators of Ne have been developed in the literature and each one uses different kinds of data (pedigree, molecular markers). But the various methods give very different estimates of Ne for the same population. Therefore, there is a need for an assessment of the consistency and reliability of the methods. The aim of our study is to analyze a few of them, making a comparison by using simulation data. A forward-in-time simulation of a multi-allelic population subjected to genetic drift was developed. We modelled a diploid population which reproduced randomly in discrete generations, with constant population size N. Self-fertilization was allowed. Selection, migration, and new mutations were assumed to not happen across generations. At each generation, Ne was estimated with various methods. We performed 700 replicates. We used the classical estimator 1/(2 x rate of Inbreeding). The coefficients of inbreeding were obtained by several ways: (i) the average pairwise coancestry derived from pedigree, (ii) the heterozygosity (He) computed from the observed allele frequencies. We also included in the comparison two estimators based on temporal changes in the allele frequency taken from the population at two points in time. We performed the comparisons varying different conditions: number of generations between sampled populations (d), initial allele configuration, and N. The results showed that the use of (ii) resulted in a huge range of Ne values, in some cases with extreme values as large as 1e+06. This is due to the large variation of the estimated values of He, which can lead to a bias in Ne. Increasing d reduced this variability of results and increased the accuracy of Ne estimation, but not very much. Estimations based on (i) resulted in very accurate estimates of Ne.

NEW PLOIDY LEVELS AND GENOME SIZE ESTIMATES IN WILD BETA TAXA USING FLOW CYTOMETRY

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It is widely recognized that the conservation of the Crop Wild Relatives (CWR) in their wild habitats, where they can continue to adapt and evolve along with their natural surroundings, is fundamental to ensure the continuous supply of the novel genetic material critical for future crop improvement. The genus *Beta* L. (Amaranthaceae) is an important source of crops, primarily for sugar production. Within a broader research project aiming to select appropriate wild accessions and identify priority localities in which to establish genetic reserves of the wild *Beta* species occurring in Portugal, the aim of this study was to assess the cytogenetic diversity of wild *Beta* populations. For this, a large scale sampling of natural populations of *B. vulgaris* subsp. *maritima* and *B. macrocarpa* was performed across Portugal (including also the archipelagos of the Azores and Madeira) and genome size and ploidy level estimated using flow cytometry. The analysed populations were mostly diploid, except for one populations of *B. macrocarpa* where two or three cytotypes (diploids, tetraploids and/or hexaploids) were found. Leaves of both species presented variable levels of endopolyploidy. The obtained results will be discussed within the context of interspecific hybridization and cryptic diversity, and constitute significant data for the conservation of these wild *Beta* crop relatives.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

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USE OF NEXTRAD AND RNA-SEQ TO DEVELOP A CONSERVATION GENOMIC RESOURCE FOR THE ENDANGERED RED COLOBUS MONKEY

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Red colobus monkeys are among the most endangered primates in the world due to habitat loss and hunting by humans. Genomic studies of these animals will assist in conservation efforts by allowing for a better understanding of neutral and adaptive genetic diversity, patterns of gene flow across a landscape, potential adaptation to known pathogens, and how best to designate conservation priorities. However, genomic approaches in this species have been hampered due to the difficulties inherent in marker development in endangered species, such as the lack of a reference genome and high quality starting biomaterials. To circumvent these issues, we applied two reduced representation genomic sequencing methods to field-collected blood samples from habituated individuals at Kibale National Park, Uganda. NextRAD, a newly developed version of RAD-seq, was employed in 10 individuals in order to develop a population-based SNP panel. We obtained between 8 - 24 million reads per sample

and identified 10,000 SNPs after removing repetitive sequences and low quality reads. RNA-seq was used to develop a set of candidate immune-related loci that can be connected to epidemiological research being conducted in this population. We performed *de novo* assembly of blood transcriptomes from four individuals and obtained a total of 18,415 contigs, with an N50 of 591-bp and a mean length of 517-bp. Fifty-two percent of the contigs BLASTed to metazoan sequences, and after mapping and annotation we identified 1,085 transcripts involved in immune responses. Ongoing research projects are using these data in this population to identify how neutral diversity relates to adaptive diversity, how habitat fragmentation affects migration and retroviral transmission, and if patterns of infection are related to immunogenetic responses. These data represent a major advance in the genomic resources available for this endangered species and will ultimately aid in efforts to help prevent their extinction.

SPECIES-GENETIC DIVERSITY CORRELATION IN ODONATE COMMUNITIES

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Similarities in the theories of community ecology and population genetics suggest that species diversity and genetic diversity within populations are shaped by the same mechanisms (selection, drift and dispersal). Thus, if the characteristics of the locality (environmental heterogeneity, community size and connectivity) influence these mechanisms in parallel on both levels of biodiversity, a positive speciesgenetic diversity correlation (SGDC) can be observed. However, also negative SGDCs are possible, if the characteristics of the locality influence the mechanisms such that they act in opposing directions on the two levels. As large scale mapping of genetic diversity is not always feasible, studies on the SGDCs can yield information of whether species diversity patterns could be used in predicting genetic diversities of populations. Also, studies on the simultaneous effect of locality characteristics on species diversity and genetic diversity can help predicting the outcomes of different management and conservation actions on both levels of biodiversity. We studied the relationships between species richness of dragonflies and damselflies, the microsatellite allelic richness of two damselfly species (Calopteryx virgo and C. splendens), and locality characteristics of their stream habitats (environmental heterogeneity, density of individuals, latitude, and connectivity to similar localities). Our analyses revealed a positive correlation between species richness and *C. virgo* allelic richness with a parallel effect of latitude being a likely explanation for the positive SGDC. However, in C. splendens, no SGDC was observed. The absence of an SGDC was likely due to species richness and allelic richness of *C. splendens* having relationships of opposing directions with connectivity to similar localities. The differing SGDCs even within closely related species highlight the variability of SGDCs in nature.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

D21SY27PS1194

IDENTIFYING THE GENES OF ADAPTATION: EXTENSIVE TRANSCRIPTIONAL PLASTICITY TO TEMPERATURE STRESS IN SISTER FRESHWATER FISH SPECIES

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Identifying genes of adaptive significance in a changing environment is a major focus of ecological genomics. Such efforts were restricted, until recently, to researchers studying a small group of model organisms or closely related taxa. With the advent of next generation sequencing (NGS), genomes and transcriptomes of virtually any species are now available for studies of adaptive evolution. We experimentally manipulated temperature conditions for two species of Australian rainbowfish (Melanotaenia duboulayi and M. fluviatilis) and measured differences in RNA transcription between control and high temperature groups. These nonmigratory species are found across a latitudinal thermal gradient in nonoverlapping areas of eastern Australia and are predicted to be negatively impacted by ongoing environmental and climatic change. Using next generation RNAseq technologies on an Illumina HiSeq2000 platform, we assembled a de novo transcriptome, mapped reads back to this transcriptome and tested for differential expression across the treatment groups. Quality of the assembly was high with a N50 length of 1856 bases. Of the 107,749 assembled contigs, we identified 4251 that were differentially expressed according to a consensus of four different mapping and significance testing approaches. Once duplicate isoforms were removed, we were able to annotate 614 up-regulated transfrags and 349 that showed reduced expression in the higher temperature group. Annotated blast matches reveal that differentially expressed genes correspond to critical metabolic pathways previously shown to be important for temperature tolerance in other fish species. Interestingly, the two sister species returned a similar list of differentially expressed genes. Our results indicate that rainbowfish exhibit predictable plastic regulatory responses to temperature stress and the genes we identified provide excellent candidates for investigations of population adaptation to increasing temperatures.

LIFE HISTORY TRADE-OFFS AND HUMAN-DRIVEN MICROEVOLUTION IN MARITIME PINE, A MANAGED FOREST TREE

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Trees are massive organisms, with great maintenance costs, long lifespan, delayed reproduction and long life cycles. Just as any other organism on Earth, they must cope with different environments by finely tuning their relative allocation of resources to growth, reproduction and maintenance. Forests -and hence forest trees- are facing new challenges due to climate warming and other human-derived impacts like forest fires. Under this scenario, selection for earlier and enhanced reproduction is predicted by life history theory. Importantly, forest management can also modify and even counteract adaptive selective pressures in natural, largely undomesticated populations. For example, traditional breeding for high timber yield is predicted to delay reproduction, and lower resistance to biotic or abiotic stressors. In spite of the sound scientific background on life history theory, forest management has largely overlooked these biological principles. Here, we review the existing evidence for how breeding interferes on microevolutionary processes of forest trees. We also provide novel examples focusing on a Mediterranean pine (Pinus pinaster), a species for which detailed quantitative genetic information for key life history traits (growth, size at reproduction, investment in chemical defences) is available. We will present two practical examples derived from breeding programs to illustrate indirect impacts of one event of artificial selection for growth already found in the following generation: first, a correlated genetic response of increased size at first reproduction and reduced reproductive allocation; and second, a reduced ability to cope a harmful forest pest. These evidences suggest that current forest tree breeding can counteract natural selective pressures, thus compromising the resilience of forest populations to future challenges. On the other hand, these finding support that assisted micro-evolution can be considered in dynamic genetic conservation programs.

27. Evolutionary Conservation: the Applied Side of Evolutionary Biology

D21SY27PS1205

OUTBREEDING DEPRESSION AND HETEROSIS ARE PREDICTED BY DEMOGRAPHY, ENVIRONMENTAL AND CYTOGENETIC CONTEXTS IN NATURAL POPULATIONS

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When individuals from separate populations mate, the resulting hybrid offspring can experience fitness benefits (heterosis) or costs (outbreeding depression) relative to their parents. Understanding the distribution and extent of these genetic benefits and costs is essential for guiding conservation plans that seek to mix wildlife populations. We use meta-analysis to test the hypothesis that phenotypic responses to intraspecific outbreeding can be predicted by combining information on population demography, environmental and cytogenetic contexts. Our dataset comprised 510 effect sizes describing intrinsic outbreeding responses, from 98 studies on animals and plants (79 species). Our results indicate that information on population context can be used to predict both observed outbreeding depression and heterosis. Studies for which we predicted a risk of outbreeding depression showed a cost to fitness (viability, survival, reproduction) in the F2 generation, relative to mid-parent performance. In cases predicted to exhibit heterosis we observed a corresponding fitness benefit relative to the mid-parent. We discuss whether and how these results, and our approach to predicting outbreeding responses, might be useful in conservation practice.

INTROGRESSION LEVELS OF THE ITALIAN AND CARNIOLAN HONEY BEE SUBSPECIES INTO THE BLACK HONEY BEE: A COMPARISON BETWEEN MICROSATELLITE AND SINGLE NUCLEOTIDE POLYMORPHISM (SNP) MARKERS

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Human activities have been shaping the distribution of honey bee subspecies in Europe. In fact, during the last decades there has been an extensive introduction of the beekeepers' favorite eastern European (lineage C) subspecies A. m. ligustica (Italian) and A. m. carnica (carniolan) into western Europe. Because of these introductions, there has been gene flow, and even replacement, of the native western European subspecies A. m. mellifera (black honey bee), which belongs to lineage M. Assessing levels of introgression is an important activity in breeding programs, especially when conservation of native subspecies is a major concern. Previous surveys of the A. m. mellifera populations estimated the introgression of lineage C into lineage M honey bees by using mtDNA and microsatellite markers. Others markers, such as SNPs, have some advantages as they provide a genome wide coverage, higher quality data, and at the same time they are suitable for automatic and standardization in high throughput technologies. Previous studies indicate that the discriminatory power of SNPs to detect population structure is lower than that of microsatellites; about 100 SNPs are needed to provide the same power of 10-20 microsatellites. In this study we will compare introgression levels between microsatellites and SNPs as estimated by the software STRUCTURE using the admixture model and correlated allele frequencies in a black honey bee collection originating from several countries across western Europe. This collection was genotyped for 12 microsatellites and 1183 SNPs. The introgression levels were first estimated using both full datasets. Then, to have similar discriminatory power between the 12 microsatellite and SNPs, we used between 60 and 120 SNP loci from the initial 1183 SNP dataset by selecting SNPs located nearby the microsatellite loci. With this work we want to verify which is the best strategy for assessing the levels of introgression in honey bee breeding programs.

SPATIO-TEMPORAL DYNAMICS OF GENETIC VARIATION AND EFFECTIVE POPULATION SIZE IN FRAGMENTED HOUSE SPARROW POPULATIONS

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Habitat fragmentation is a major threat to biodiversity: by reducing population sizes and gene flow between populations it affects genetic structure and effective population size (*Ne*), which have important implications for population dynamics and evolutionary processes. We used natural house sparrow (*Passer domesticus*) populations in Norway as a model system to investigate if population type (i.e. mainland or island), geographic distance, adult population size (*N*), immigration rate and sex ratio explained intra- and interpopulation genetic variation and *Ne* of fragmented populations. Data from microsatellite genotyping across multiple populations and generations was used. Our results showed that intrapopulation genetic variation was lower and the occurrence of population bottlenecks more frequent on islands than the mainland, and the general level of genetic differentiation was higher between islands than between mainland populations, but only at shorter distances. Furthermore, genetic differentiation decreased, whereas intrapopulation genetic variation and Ne/N-ratios increased with immigration rates. Both intrapopulation genetic variation and Ne increased with population size. However, genetic Ne was much larger than demographic *Ne* (often even >N), probably due to a greater effect of immigration on genetic than on demographic processes. In constrast, genetic Ne for the metapopulation were within the expected range (*<N*), suggesting that in fragmented populations even low levels of gene flow may make the total metapopulation the appropriate scale of estimation. We also developed a genome-wide 10 000 Single Nucleotide Polymorphism (SNP) chip for the house sparrow and screened individuals from four populations. This demonstrated that genome-wide marker data gave results similar to microsatellites. Our results are relevant for a better understanding of evolutionary processes and hence conservation of threatened populations.

SMALL SCALE GENETIC STRUCTURING IN THE NEOTROPICAL PALM TREE EUTERPE EDULIS (ARECACEAE)

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The Atlantic Rainforest extends along the Brazilian coast covering a wide latitudinal range with great variations in altitude. Most studies of its species genetic variability focus on a large scale sampling design, but small scale studies are also important as they provide insights about local processes. Euterpe edulis is an endangered neotropical palm, distributed throughout the Atlantic Forest from the coastal plain up to 1000 meters above sea level. We evaluated the genetic diversity and structure in six natural populations of *E. edulis* from different altitudes in a continuous protected area at the north coast of São Paulo State; 50 adults from each location were genotyped with seven microsatellite loci. The total allele number was high (140) and the mean allele number did not vary between samples. The total expected heterozygosity was 0.867 ranging from 0.782 to 0.859 among samples. The inbreeding coefficient was low in all samples, as expected for outcrossing species. The spatial genetic structure was absent or weak at populations level. The genetic structure was high ($\theta' = 0.26$), considering that the maximum distance between samples was 32 Km. Four most likely genetic groups were defined by the assignment test, and five according to AMOVA. A partial Mantel test correlated the pairwise genetic structure with the geographical distance (r = 0.8; p < 0.05) and also with the pairwise altitudinal differences without the effect of the geographic distances (r = 0.5; p < 0.05). Those differences may be due to reduced gene flow or local adaptation, what remains to be studied as potentially relevant for management programs. Such a pattern of genetic differentiation at short distances is unexpected within a continuous rainforest in a protected area, highlighting the importance of small scale approaches to better understand the complex patterns of differentiation in tropical systems.

GENOMIC CONSEQUENCES OF INBREEDING AND HYBRIDIZATION IN PIGS AND WILD BOARS

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The maintenance of genetic diversity in (isolated) populations is essential for their adaptive ability. If standing variation is low, the susceptibility to a variety of diseases and environmental changes is elevated. Two haplotypes are IBD (identical by descent) when they originate from a common ancestor. If IBD haplotypes reunite within one diploid organism, it results in a region of homozygosity (ROH), which is an indication of recent inbreeding. The interplay of population demographic history and recombination rate is essential in the process of haplotype formation and distribution. In management of small, potential inbreed, populations, outcrossing is used to introduce new haplotypes in a population and hereby increase genetic diversity. This study utilizes the latest genomics tools to identify shared haplotype tracts between pigs (Sus scrofa) from different Eurasian wild boar and breed populations. Pigs are an excellent model species to study haplotype structure because of their complex demographic history, multiple domestication events and recent admixture. Next-generation sequencing enables a thorough and nearly ascertainment bias-free investigation of genome-wide patterns of haplotype sharing. Our study reveals patterns of inbreeding, and conserved, introgressed and selected haplotypes in the pig genome. Haplotypes that are shared between closely related individuals are longer and more abundant in the genome than haplotypes shared by distantly related individuals. Selection on introgressed haplotypes results in an over-representation of these haplotypes in a population. Nucleotide diversity in the genome is higher when at least one haplotype is shared with a distantly related individual, supporting the fundamental genetics theory behind outcrossing. Detailed mapping of the genomic distribution of variation enables a targeted approach to increase genetic diversity of captive and wild populations, which may facilitate conservation efforts in the near future.

LIFE HISTORY TRADE-OFFS AND HUMAN-DRIVEN MICROEVOLUTION IN MARITIME PINE, A MANAGED FOREST TREE

Jose Climent¹, Luis Santos del Blanco², Luis Sampedro⁴, Zas Rafael³, Alía Ricardo¹ ¹Population Ecology and Evolution, Forest Ecology and Genetics, INIA-CIFOR, Spain ²Ecology and Evolution, University of Lausanne, Switzerland ³Forest Genetics Lab, Misión Biológica de Galicia-CSIC, Spain ⁴Forest Genetics Lab, Ecology, Centro de Investigación Forestal de Lourizán, Spain climent@inia.es

Trees are massive organisms, with great maintenance costs, long lifespan, delayed reproduction and long life cycles. Just as any other organism on Earth, they must cope with different environments by finely tuning their relative allocation of resources to growth, reproduction and maintenance. Forests -and hence forest trees- are facing new challenges due to climate warming and other human-derived impacts like forest fires. Under this scenario, selection for earlier and enhanced reproduction is predicted by life history theory. Importantly, forest management can also modify and even counteract adaptive selective pressures in natural, largely undomesticated populations. For example, traditional breeding for high timber yield is predicted to delay reproduction, and lower resistance to biotic or abiotic stressors. In spite of the sound scientific background on life history theory, forest management has largely overlooked these biological principles. Here, we review the existing evidence for how breeding interferes on micro-evolutionary processes of forest trees. We also provide novel examples focusing on a Mediterranean pine (*Pinus pinaster*), for which detailed quantitative genetic information for key life history traits (growth, size at reproduction, investment in chemical defences) is available. We will present two practical examples derived from breeding programs to illustrate indirect impacts of one event of artificial selection for growth already found in the following generation: first, a correlated genetic response of increased size at first reproduction and reduced reproductive allocation; and second, a reduced ability to cope a harmful forest pest. These evidences suggest that current forest tree breeding can counteract natural selective pressures, thus compromising the resilience of forest populations to future challenges. On the other hand, these findings support that assisted microevolution can be considered in dynamic genetic conservation programs.

POPULATION STRUCTURE AND GENE FLOW OF DOGWHELK (NUCELLA LAPILLUS) IN WESTERN SCOTLAND AND ENGLAND

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Population structure and gene flow of dogwhelk (Nucella lapillus) in Western Scotland and England. Ratima Karuwancharoen, Martyn Lucas& A.Rus Hoelzel School of Biological and Biomedical Sciences Durham University, Durham, DH1 3LE, UK Email: ratima.karuwancharoen@durham.ac.uk Dog whelks are predatory marine snails which are found around rocky intertidal zones of the North Atlantic. Regional dog whelk populations are known to have declined due to tributyltin pollution during the 1970s and 1980s. This was, however, followed by successful recolonisation of this species after 1987. They lack planktonic larval stages and develop into crawling juveniles which emerge from capsules. This life history implies limited dispersal ability. However, recent research has identified long-distance dispersal in dog whelks in the eastern North Atlantic, most likely associated with 'rafting'. Some populations of dog whelks are also apparently differentiated according to the degree of wave impact on rocky shores. These findings illustrate the need for further research, especially in the context of ongoing impact from pollution and other factors. In this study we consider the relative importance of geographic distance, local habitat characteristics and discontinuous habitat in the assessment of gene flow among coastal sites in northeastern England. Most of these sites are designated protected areas. Initial results based on comparisons among 6 locations at 14 microsatellite DNA loci suggest fine-scale population structure in this system.

Symposium

28. Non-Genetic Transfer of Immunity Across Generations – Evolution and Underlying Mechanisms

21 August



Program

Wednesday 21 August

Session(s): 7, 8

Organisers: Olivia Roth and Gerrit Joop

Invited speakers: Mike Boots

Description:

To cope with parasites and pathogens, immunological experience can be transferred across generations also non- genetically, allowing for higher adaptive plasticity. Recent studies suggest that transgenerational immune priming (TGIP) can also be provided by innate defence components and that it is not limited to maternal effects. Both evolutionary and mechanistic advances in TGIP should be addressed in this symposium in systems ranging from invertebrates to vertebrates.

D21SY28IT17:45R9

EPIDEMIOLOGICAL AND EVOLUTIONARY IMPLICATIONS OF IMMUNE PRIMING

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The body of empirical evidence for immune priming in invertebrates is growing. To highlight this, here, I will give an example of within and transgenerational priming in an insect to challenge by a natural DNA virus. Given this widespread phenomena and the fact that priming, defined here as immunity following challenge to a pathogen, contrasts epidemiologically with acquired immunity which occurs after recovery from an infectious state, it is important to model the implications of priming. Defined in this way priming can also be a characteristic of vertebrate immunity. Here I will describe the epidemiological and evolutionary characteristics of a SPI (susceptible:primed:infectious) model as distinct from the SIR (susceptible:infected:recovered/removed) model of acquired immunity. The models highlight the potential importance of priming to the epidemiology and evolution of host parasite interactions.

D21SY28RT15:45R9

IMMUNE-BASED MATERNAL EFFECTS IN RESPONSE TO EXPERIMENTAL MANIPULATION OF BACTERIAL LOAD IN TREE SWALLOW NESTS

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Birds encounter a variety of bacteria in their environment on a daily basis. The vast majority of these organisms likely exert minimal direct effects on avian fitness, while others may exert negative pathogenic effects or conversely, positive probiotic effects. Regardless of pathogenicity, the antigenic properties of many bacteria likely contribute to variation in immune status among individual birds at any given time. Some of this variation may in turn be inherited from mother to offspring through the deposition of protective immune compounds into eggs. Neonates may be especially sensitive to environmental bacteria during the first weeks of life, before the immune system is fully developed and the gut microbiota established. And thus, variation in female exposure to bacteria prior to egg-laying may result in indirect fitness consequences through maternal effects on early offspring immune phenotype. In this study, I manipulated bacterial load in tree swallow nests during nest-building and egg-laying to explore bacterial effects on female investment in egg defenses and on offspring growth and immune status. Contrary to my predictions, eggs collected from nestboxes with added bacteria contained lower concentrations and total amounts of yolk antibodies than eggs from control nests. These patterns, observed for total yolk antibodies, were also reflected in nestling plasma samples. Nestlings whose mothers were exposed to increased bacterial load also exhibited lower bactericidal capacity than control nestlings at six days post-hatch, but not at twelve days post-hatch. No differences in nestling size, growth, or PHA responsiveness were observed among treatment groups.

D21SY28RT16:09R9

CONTEXT-DEPENDENT PATERNAL EFFECTS OF INFECTION ON OFFSPRING SURVIVAL AND CONDITION

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Besides traditional genetic inheritance, the phenotype of an individual can be influenced by its parents' phenotypes and environment. Whether those environmentally-induced parental effects are adaptive though remains elusive. To address this question, we focused on paternal effects of exposure to a nematode parasite in the three-spined stickleback. We experimentally investigated how parasite infection affects adult stickleback males and their offspring's early life history traits and parasite resistance. By means of *in vitro* fertilization, we produced over 50 maternal half-sibships sired by pairs of infected and uninfected brothers. We first present evidence for sperm deficiency in infected fathers compared to uninfected fathers (reduced velocity and fertilizing ability). We then show that although offspring of infected fathers suffered from low hatching success and survival rates, when exposed to the same parasite, they achieved higher body condition than their counterparts from uninfected fathers. These results highlight that the adaptive potential for paternal effects is only expressed in the presence of the paternal environmental stressor. We thus discuss the likelihood and potential consequences of sperm-mediated transgenerational effects under stable or predictable environments.

D21SY28RT16:33R9

BI-PARENTAL IMMUNE PRIMING IN THE SEX-ROLE REVERSED PIPEFISH SYNGNATHUS TYPHLE

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The transfer of immunity from mothers to offspring (trans-generational immune priming) yields in an improved offspring immunity and an induced parasite resistance. In teleost this occurs via maternal deposition of antibodies into the egg yolk. As yet, the paternal contribution to offspring immunity was suggested to be negligible because sperm are too small for a transfer over the scope of the DNA. Syngnathids like the sex-role reversed pipefish Syngnathus typhle have evolved a paternal placenta-like structure, in which embryos are nursed. This gives a mechanistic opportunity for paternal transfer of immunity but also implies that offspring are born in the paternal environment and thus share a similar parasite pressure. Hence, strong selection for the evolution of paternal immune priming can be expected.
We experimentally investigated the potential of maternal, paternal and bi-parental immune priming on offspring cell activity and gene expression patterns. We found that offspring's immune defence was enhanced upon parental bacteria exposure, independent of whether mother, father or both were challenged. This suggests that male pregnancy let to the evolution of bi-parental immune priming. Classically, maternal immune priming is considered to be of higher importance due to the fact that mothers invest more resources in production and care of offspring. However, since in sex-role reversal males do not only have an intense paternal investment but also realize evolutionary maternal traits like a more efficient immune response, selection pressures for paternal immune priming should be stronger. As immune priming is expected to be energetically costly for both parents and offspring, life history parameters provide clarification about incorporated costs and benefits. Further, future data will give insights in the mechanistic basis of bi-parental immune priming and assess what role antibodies, innate immunity and epigenetics play.

D21SY28RT16:57R9

TRANSGENERATIONAL IMMUNE PRIMING IN LEPIDOPTERA

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Parental investment in offspring encompasses its preparation to defend themselves against pathogens or parasites initially encountered by the adults. In order to explore the mechanisms mediating transgenerational immune priming in insects we used the established lepidopteran model host Galleria mellonella. Here we show that feeding of G. mellonella larvae with either non-pathogenic bacteria or with bacterial pathogens such as Pseudomonas entomophila and Serratia entomophila is sufficient to increase expression of immunity-related genes such as the antibacterial protein gloverin. Dietary uptake of bacteria by the larvae resulted also in differential expression of immunity-related genes in the eggs laid by the corresponding female adults, providing evidence for trans-generational immune priming of the offspring. In subsequent studies we explored the mechanism mediating transgenerational immune priming in Lepidoptera.

D21SY28RT18:33R9

EVOLUTION OF A TRANSGENERATIONAL RESPONSE TO PARASITES, THE MATERNAL TRANSFER OF ANTIBODIES: THEORY, EXPERIMENTS AND COMPARATIVE APPROACHES

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Among the many responses that hosts show against parasites, the transgenerational transfer of immunity is now receiving increased attention due to its broad potential evolutionary, epidemiological and ecological implications. In vertebrates, the maternal transfer of antibodies is a transgenerational response to parasites for which there is a relatively strong mechanistic and biomedical understanding of the processes involved. Recent theoretical work and experiments we conducted to address evolutionary predictions are nevertheless shedding new light on both the proximate and ultimate processes involved. We notably found that the temporal persistence of maternal antibodies in young birds after hatching can be much longer than previously thought, and that this persistence is predictably linked to the life history of the considered species. Because of the mechanistic link between antibody persistence in adults and young, this has implication for the evolution of acquired immunity. We also showed that offspring can receive maternal antibodies via allosuckling in social mammals. Such results have wide implications, from comparative immunology and the evolution of social behaviours, to the use of vaccines as management tools in eco-epidemiology. They also call for further work at the interface between mechanistic and evolutionary approaches.

D21SY28RT18:57R9

MATERNAL IMMUNE CHALLENGES EXERT BOTH ENHANCING AND SUPPRESSIVE EFFECTS ON OFFSPRING IMMUNE RESPONSES

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Maternal antibodies can both enhance and suppress offspring humoral immune responses. However, these enhancing and suppressive effects have typically been documented in separate studies conducted with different species and different antigens over variable time periods. Thus the conditions that induce a priming effect of maternal immune challenge versus a suppressive effect have been unclear. In this study, we exposed female zebra finches (Taeniopygia guttata) and their offspring to one of two antigens (keyhole limpet hemocyanin (KLH) or lipopolysaccharide (LPS)) or a control treatment and quantified maternal antibody transmission and endogenous offspring antibody production. Antigenspecific maternal antibodies reached their lowest levels between days 17 and 28 post-hatch and total antibody concentrations peaked at day 17 post-hatch. We found that both maternal KLH and maternal LPS exposure stimulated the production of antigen-specific antibodies in non-challenged offspring after the catabolism of maternal antibodies. This suggests a stimulatory effect of maternal immune challenge on offspring humoral immunity. However, offspring of both KLH and LPS challenged mothers exhibited weaker antigen-specific secondary immune responses when mothers and offspring were challenged with the same antigen. Maternal immune challenge did not impact the ability of young to respond to the other antigen. This supports a specific, suppressive effect of maternal immune challenge on offspring antibody production. Thus, maternal antibodies can have an instructive role, potentially acting as internal antigens, and stimulate offspring antigen-specific antibody production in response to two distinct antigens. However, when the early antigenic environment of offspring is similar to the maternal antigenic environment, maternal antibodies may suppress the endogenous antibody responses of young.

POSTERS

D21SY28PS0084

THE TEMPORAL DYNAMICS OF IMMUNE GENE EXPRESSION AND PATHOGEN LOAD REVEAL RESISTANCE AND TOLERANCE OF BACTERIA INFECTION BY TRANS-GENERATIONALLY PRIMED FLOUR BEETLES

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Immune priming allows individuals previously exposed to a pathogen to enjoy reduced susceptibility and higher survival probability upon re-exposure, compared to naïve cohorts. The protection can even be transferred across generations, but currently very little is known about the mechanisms that confer a primed immune response in invertebrates. Furthermore, whether trans-generational immune priming increases resistance (ability to kill pathogens), tolerance (ability to minimize or repair damage), or both is unknown. In trans-generational priming experiments reported here, we infected the larval offspring of naïve, sterilely wounded, or bacteria-challenged (primed) adult female beetles (Tribolium castaneum) with the bacterial entomopathogen Bacillus thuringiensis (Bt) and used DNA microarrays and qPCR to investigate the influence of maternal treatment on the temporal dynamics of bacterial load and immune gene expression. Meanwhile, we monitored the survival and development of infected and uninfected larvae from these treatments to quantify the functional outcomes of priming in this system. Initially, primed larvae were better able to curb bacterial proliferation and they subsequently exhibited increased tolerance for high bacterial loads. Moreover, primed larvae had better survival odds and buffered developmental costs of infection. Taken together, these results suggest that invertebrate immune priming is not just an outcome of more efficient microbe killing, as previously supposed, but rather a two-pronged reduction in the overall negative consequences of infection for the host, with broad implications for the evolution of immune priming and pathogen virulence strategies.

D21SY28PS0272

ORAL IMMUNE PRIMING IN THE RED FLOUR BEETLE TRIBOLIUM CASTANEUM

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The phenomenon of immune priming and its transfer to the offspring as trans-generational priming has now been demonstrated for number of insect species, including the red flour beetle Tribolium castaneum. For testing this phenomenon, the majority of studies have focused on introducing the pathogen into the insects' hemolymph by making a small wound in the cuticle. Although such septic injury can occur in nature, many pathogens are able to infect their hosts also via the oral route, i.e. by uptake with the food. Bacillus thuringiensis is a spore-forming, entomopathogenic bacterium, which infects its hosts in this way, and penetrates the host gut epithelium to gain access to the hemocoel. We tested for immune priming of T. castaneum larvae by oral exposure to B. thuringiensis vegetative cells and spores, as well as by media supernatants from bacterial cultures. We found strong induction of immune priming when we exposed the larvae to the sterile media in which bacteria were raised, but not to the heat-killed bacteria, killed spores or low amounts of viable spores. Primed larvae showed increased survival upon oral challenge with a high dose of *B. thuringiensis* spores. Moreover, they had a longer developmental time, indicating that investment in such a priming response comes with a cost. We moreover show that increased survival in larvae exposed to the priming-inducing diet was not caused by a difference in the size of larvae after priming. We are currently testing for trans-generational oral priming, i.e. whether the offspring of primed individuals shows increased survival upon challenge.

D21SY28PS0409

MATERNAL INVESTMENT AND LARVAL FITNESS IN THE BURYING BEETLE NICROPHORUS VESPILLOIDES

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Studies of transgenerational immune priming have shown that an immune challenge to parents can lead to upregulated offspring immunity and increased resistance to disease1,2. Here we investigate whether mothers can anticipate immune challenges to their offspring by assessing the quality of resources that will be available to offspring. When resources are spatially and temporally variable, it could be adaptive to alter the offspring phenotype accordingly. The burying beetle Nicrophorus vespilloides feeds and reproduces on carrion. Females will breed multiple times during their lives, and carcasses will vary in levels of decomposition throughout their lifespan. Their offspring will therefore experience different levels of competition and challenges from micro-organisms, as well as variation in nutritional quality. Parents examine and carefully prepare carcasses for their broods, giving mothers an opportunity to assess the quality of the reproductive resource during this preparation phase. We tested the hypothesis that beetle mothers can anticipate the offspring's environment during the preparation of the resource and match offspring phenotype to this by adjusting immune investment and/or development. We performed a cross-factorial experiment in which mothers bred on resources of either good or poor quality, and newly hatched larvae were transferred onto resources of either the same or different quality. During their final larval instar, traits relating to fitness and immune defence were measured. Adult life history traits from the offspring generation were also assessed, and we present those findings here. References: 1 Tidbury, H., Pedersen, A., Boots, M. 2011 Within and transgenerational immune priming in an insect to a DNA virus Proc. R. Soc. B 278, 871-876 2 Sadd, B.M., Kleinlogel, Y., Schmid-Hempel, R. & Schmid- Hempel, P. 2005 Trans-generational immune priming in a social insect. Biol. Lett 1, 386–388

D21SY28PS0431

PRIMING AGAINST PATHOGEN ATTACK IN CAENORHABDITIS ELEGANS – EFFECT OF PROBIOTICS OR IMMUNE SYSTEM ACTIVATION?

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Parasites impose strong selective pressures on hosts since they reduce per definition host fitness. Immune reactions are advantageous, but they are also costly. Therefore, mechanisms reducing the costs of a continuous expression of immune genes are selected. An alternative strategy is the behavioural defence, for example escape behaviour. Hosts prevent contact to parasites by escaping potentially dangerous areas, particles or individuals. For the nematode *Caenorhabditis elegans* such escape behaviour is well known: It escapes pathogenic bacteria and prefers feeding on non-pathogenic variants of the same bacterial species. We here ask whether feeding on these non-pathogenic bacterial variants offers another advantage than just the uptake of non-pathogenic food. In detail we test whether previous contact to non-pathogenic *Bacillus thuringiensis* is advantageous upon later contact to pathogenic bacteria survive better upon contact to pathogenic bacteria. Since this effect is equally strong if the non-pathogenic bacteria are alive or dead, this effect is most likely caused by an immune activation and not by the probiotic accumulation of non-pathogenic bacteria in the gut. These results highlight that also in species with a primitive immune system immune priming may occur, also the exact mechanism remains unclear.

D21SY28PS0581

FOSTER CARE-GIVERS INFLUENCE BROOD PATHOGEN RESISTANCE IN ANTS

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Social organisms face a high risk of epidemics, and respond to this threat by combining efficient individual and collective defenses against pathogens. An intriguing and little studied feature of social animals is that individual pathogen resistance may depend not only on genetic or maternal factors, but also on the social environment during development. Here, we used a cross-fostering experiment to investigate whether the pathogen resistance of individual ant workers was influenced by their care-givers' or their own colony of origin. The ability of newly eclosed, cross-fostered *Formica selysi* workers to resist the fungal entomopathogen *Beauveria bassiana* was influenced by the workers that cared for them during development. This effect persisted even in the absence of ongoing social interactions and did not depend upon the social structure of the colony of origin of either care-givers or brood. Social interactions during development thus influence individual resistance, blurring the line between individual and social immunity.

D21SY28PS1008

THE ROLE OF MATERNAL EFFECTS ON THE ONTOGENY OF CONSTITUTIVE IMMUNITY IN WILD BIRDS

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The ability to resist infections early in life is expected to be under intense selection pressure because infection may have severe impact on fitness. Maternally transmitted immune components provide some protection during early stages and may have long-lasting effects on the development of the humoral immune system. Extrinsic influences on mothers (i.e. exposure to pathogens) may also influence vertical transmission of immune components to offspring. However, maternal effects on the ontogeny of offspring immune function may not be restricted to vertically transmitted components. Mothers may influence the development of offspring immune function by providing a suitable environment for embryo development through parental care. Indeed, parental behaviour in birds plays an important role in keeping embryo thermal conditions within the optimal range for development. This parentally induced variation in embryo temperatures can thereby influence development and performance of the immune system. Yet, whether these maternal effects mediated by egg constituents or thermal environment interact with endogenous development of immune function differently among species remains unstudied. In this study, we examined the ontogeny of constitutive immunity in relation to developmental stage in a group of 22 Passerine bird species. Our results show that inter-specific variation in immune activity at hatching was mainly explained by extrinsic factors mediated by the mother, suggesting an important role of maternal effects on offspring immunity at hatching. Activity of constitutive components of the immune defense was detected as early as 1-3 days post-hatching, and increasing with age, indicating that immune function in older nestlings reflects intrinsic development. Our results highlight both endogenous immune activity of altricial nestlings at an early developmental stage, and maternal effects on the ontogeny of immune function of young birds.

D21SY28PS1088

CHANGES IN FEEDING RATE CAN EXPLAIN THE MATERNAL EFFECT OF FOOD ON DISEASE RESISTANCE IN DAPHNIA MAGNA

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Maternal effects are increasingly recognised to impact on infectious disease. In invertebrates, maternal parasite exposure, as well as changes in maternal abiotic environment (e.g. food availability, crowding, forced-flight), have been shown to influence the outcome of offspring-parasite interactions with profound fitness consequences for the host. An understanding of the mechanisms underlying these maternal effects is a prerequisite for recognising their adaptive value, yet proximate mechanisms have so far received little attention. We investigated the mechanism underlying a maternal effect on disease resistance in the crustacean *Daphnia magna*, an organism that produces offspring more resistant to the sterilising bacterium *Pasteuria ramosa* when food availability is relatively poor. We show that this maternal effect is likely to be mediated by a reduction in feeding rate in the offspring of food stressed mothers, explaining their improved resistance to a parasite that infects via the gut.

D21SY28PS1263

TRANSCRIPTOME ANALYSIS OF IMMUNE PRIMING IN THE BUMBLEBEE (BOMBUS TERRESTRIS)

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The innate immune system of invertebrates has been claimed to be unspecifically acting against saprophytes based on the observation, that the genes contributing to innate immunity are well conserved across species. However, in recent years specificity and a form of memory in invertebrate immunity (immune priming) have been shown to occur in a number of species. Homologous infections led to higher survival than heterologous infections. Especially long lived species that might be at risk of subsequent infections might have a selective advantage when developing a potentially costly immune response that shows specificity and longevity. As the underlying mechanisms of immune priming are not properly examined and understood we used a transcriptome analysis of heterologous and homologous infections using gram-positive and -negative bacteria in the bumblebee, Bombus terrestris. The results indicate that the gene expression pattern are determined by initial infections rather than by secondary pathogen exposure. Thus, the immune system is primed by the first pathogen to be encountered and shows a long-lasting response at the level of gene expression. Effector genes of the innate immune system pathways (e.g. antimicrobial peptides), which are differentially activated by gram-positive and -negative bacteria, do show a specific, long-lasting response initiated by the initial infections. Using a transcriptomic approach we are able to unravel the mechanism of immune priming in insects. Initial infections are indeed priming the immune response so that a secondary homologous infection might be better counteracted due to specifically expressed genes of the immune system than a heterologous infection.

D21SY28PS1296

AVIAN MATERNAL TRANSFER OF ANTIBODIES: ARE THEY AFFECTING THE SPECIFIC IMMUNE RESPONSE OF THEIR CHICKS?

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Avian parents influence their offspring's phenotypes by providing different sorts of parental care. Mothers may transfer a variety of elements through the yolk sack that can affect the embryo's development. Among these elements supplied, mothers transfer antibodies that provide transient adaptive immune protection to the chick. However, whether such transient specific immunity is enhancing the chicks' subsequent immune response to the same specific immune challenge remains controversial. We explored whether mother's specific immune response to Newcastle disease virus (NDV) affected their chick's subsequent response to the same antigen. We studied a wild breeding population of house sparrow (Passer domesticus) in southern Spain. We captured 57 breeding females in two consecutive breeding events: 29 were vaccinated against NDV in their first brood, and the rest were sham-controlled. Chicks on their second brood were equally treated, the concentration of NDV antibodies (NDV-Ab) was measured, and the phytohaemaggulutinin-induced immune response (PHA) assayed from all before fledging. Leukocyte profile, antioxidative status and total protein concentration from blood was measured from all individuals. Both vaccinated mothers and chicks presented higher NDV-Ab levels than control individuals. Chicks's level of NDV-Ab was marginally affected by their mother's treatment, which was further confirmed when the level of maternal NDV-Ab was taken into consideration. Chicks challenged with NDV vaccine developed a stronger specific response whenever their mothers presented high levels of NDV-Ab. However, chick's PHA response was unaffected by none of the treatments, nor the mothers' condition (NDV-Ab, leukocyte profiles or antioxidative status) in any way. Our results suggest that house sparrow mothers transfer specific NDV-Ab, which help their chicks to respond more strongly when challenged with the same antigen, leaving other immune responses unaffected.

Symposium

29. Actively Learning Evolution: Methods and Resources

20 August



Program

Tuesday 20 August

Session(s): 1

Organisers: Alexandra Isabel Sá Pinto, Rita Campos and Kristin

Jenkins

Invited speakers: Tom Meagher

Description:

This symposium aims to explore methods and resources available for inquiry based teaching of evolution and to discuss the impacts of such methods on both students' and the public's understanding and engagement with evolution. Speakers will present information about effective teaching methods and demonstrate methods and resources through hands on activities.

D20SY29IT10:30R7

DATA ACCESS AND SCIENTIFIC SOCIETIES

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As part of the changing landscape of science, data archiving is becoming a widespread practice. This has in turn created an opportunity to more directly integrate archived data resources into teaching at all levels. For example, it is possible to develop student exercises that reconstruct the underlying analysis presented in published work, and perhaps to explore other dimensions of such work in creative ways. In order to facilitate such exploration-based learning, there is a need for developing scientific data as a teaching tool. An approach to doing so being led by the Ecological Society of America, in collaboration with the Society for the Study of Evolution and other scientific societies, has been to establish an peer-reviewed data-based online resource library educational modules. EcoED of (http://ecoed.esa.org/). In the very near future, this online resource is planned to grow to include various modules. including one geared to the evolution community, EvoED (http://evoed.evolutionsociety.org/). Integration of the vast online resource into educational modules that encourage exploration and analytical approaches to science is an important step change in science teaching that will promote better understanding among students of science as a process as well as a product.

D20SY29RT11:18R7

IMPLEMENTATION OF PROBLEM-BASED LEARNING IN THE CONTEXT OF CELEBRATING THE CENTENARY OF ALFRED WEGENER'S FIRST PUBLICATION ON CONTINENTAL DRIFT AND BRIDGING THE EVOLUTION SCIENCE-BIOLOGY AND GEOLOGY TEACHING PRACTICE GAP

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Alfred Wegner proposed that since the Palaezoic the continents and oceans were not fixed and that they were grouped in a supercontinent Pangaea. This theory was proposed for the first time on 1912 at a meeting with a lecture (Jacoby, 2012). This episode of the History of Science (HS) is considered an excellent example for teaching students the Nature of Science (NOS) and the interaction of the social and tecnological context in Science. During this academic year we are implementing a NOS and HSenriched Problem-Based Learning (PBL) environment at the middle (year 7 of the Portuguese National Curriculum) and secondary (year 10) level in the context of celebrating the centenary of the first publication of this theory. The goal of providing detailed implementation practices is to adress the lack of "how to" in PBL implementation in classrooms since, as indicated in the literature, is still relatively uncommon for 12 to 14 year old students because of many clallenges and barriers (Liu et al., 2012) and is the first practical example of implementation using this episode of HS for teaching evolution. We propose resources and implementation techniques for teaching evolution under the themes "biodiversity" and "mobilism" (of the Portuguese National Curriculum in both levels). The ill-problem presented to students, without a single correct answer, was based on the phylogeny of extant and extinct ratite birds (Lieberman, 2005), the ones described by Charles Darwin and the present geographical distribution. The evaluation of the students was focused on the chain of reasoning employed; and the comparisation analysis of the solution itself by the 2 classes was made. Acknowledgments Escola Secundária Aurélia de Sousa, Porto, Portugal. References Jacoby, W. R. (2012). Geoscientist 22(9): 12-17. Lieberman, B. S. (2005) Palaeogeography, Palaeoclimatology, Palaeoecology 219: 23– 33. Liu, M. et al. (2012). The Interdisc. Journal of Problem-based Learning 6(2): 46-84

D20SY29RT11:42R7

THE JACKPROT SIMULATION: SLOT-MACHINE MODEL TO TEACH THE NON-RANDOM NATURE OF PROTEIN EVOLUTION

<u>Guillermo Paz-y-Mino-C</u>¹, Avelina Espinosa¹ ¹in-group, Biology, University of Massachusetts Dartmouth, United States ²Biology, Roger Williams University, United States gpazymino@umassd.edu

Protein evolution is not a random process. We use slot-machine probabilities and ion channels, in an inquiry-based learning scenario, to show biological directionality on molecular change. The slotmachine represents the cellular chemical apparatus, product itself of Darwinian evolution, required to generate, step by step, each of the nucleotides coding for an amino acid of a model protein. Teachers and students can access the Jackprot Simulation and run statistical analysis of protein evolution by cutting and pasting nucleotide sequences obtained from the WWW. The Jackprot generates statistics on nucleotide evolution under selection (observed vs. expected values) and at random (without selection). We will use the following example when explaining hands-on how to use the Jackprot: Because ion channels reside in the lipid bilayer of cell membranes, their residue location must be in balance with the membrane's hydrophobic/philic nature; a selective 'pore' for ion passage is located within the hydrophobic region. We will contrast the random generation of DNA sequence for KcsA, a bacterial two-transmembrane-domain (2TM) potassium channel, from Streptomyces lividans, with an underselection scenario, the 'Jackprot,' which predicts much faster evolution than chance. We will distribute guidelines on how to use the online interface The Jackprot Simulation (JAVA APPLET Version 1.0) to model a numerical interaction between mutation rate and natural selection during the scenario of polypeptide evolution. Winning the 'Jackprot,' or highest-fitness complete-peptide sequence, requires cumulative smaller 'wins' (rewarded by selection) at the first, second and third positions in each of the 161 KcsA codons ('jackdons' that led to 'jackacids' that led to the 'Jackprot'). The 'Jackprot,' as didactic tool, helps students understand how mutation rate coupled with natural selection suffice to explain the evolution of specialized, complex proteins. Student learning data will be shared.

POSTERS

THE ROLE OF CHANCE IN EVOLUTION: HOW TO TEACH IT IN THE CLASSROOM

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Comprehend the role of chance in evolutionary processes has been one of the most notorious difficulties in the practice of teaching and learning of evolution. Historical research has noted the persistence of teleological and finalist thinking even after taking courses of evolution. The proposal is based on the builder metaphor (or the architect metaphor) used by Charles Darwin to explain the causal disconecction between variation's origin and the process of natural selection (C. Darwin, 1868, The variation of animals and plants under domestication). Darwin developed this rethoric form to show the contingency (or chance) in the evolutionary history of the species, and to explain that variations that arise from natural causes, they do not have in their origin any relation to the adaptive role that variations have in the life history of organisms. This proposal is divided in three sections: First, a description of the builder metaphor; second, how the metaphor can be used to explain the role of chance in a human-made construction: a "stone snake", and third, an explanation of the role of chance in the origin and evolution of rattlesnakes (Crotalus sp), emphasizing the following characteristics: elongated body, lack of limbs, flathead distinguished from the body, and three specific characteristics of the genus Crotalus: specialized dentition or solenoglyph, sensitive pits to infrared light and the rattlesnake. This proposal is intended for teachers of upper secondary education and seeks to explain schematically trough illustrations the role of chance in evolutionary processes.

BUILDING SCIENTIFIC CAPACITY IN CAPE VERDE ISLANDS: INTEGRATING DIFFERENT APPROACHES TO STUDY THE EVOLUTION OF THE ENDEMIC FLORA

Maria M Romeiras¹, Monica Moura², Miguel M Sequeira³, Maria C Duarte¹ ¹JBT, IICT-Tropical Research Institute, Portugal ²Department of Biology, CIBIO, InBIO, Polo dos Azores, Portugal ³GBM, University of Madeira, Portugal mromeiras@yahoo.co.uk

Understanding the origin and evolution of the Cape Verde endemic flora is essential for the conservation in this Biodiversity Hotspot. A recent funding research project by FCT-Portugal, entitled "Conservation of plant biodiversity in the Macaronesian Hotspot: Integrating phylogenetic, taxonomic, and ecological approaches to study the Cape Verde endemic flora" will address different questions, namely: (a) How does an extraordinarily rich endemic flora evolved? and (b) Why is the flora so threatened and how might its conservation be secured? In this communication we show how the fieldwork activity can play an important role to help graduated students understanding and learning plant evolution; it is expected that at the end of each botanical expedition, students will be able to understand the scientific methods used for the study evolutionary processes in this archipelago. We advocate that international collaborations can build scientific capacity in developing countries and will contribute to influence in a positive way students' attitudes about biodiversity, as well as, to encourage further studies in this insular ecosystem. Moreover, by stimulating a positive attitude and providing knowledge about biodiversity and the consequences of its loss, students realize why scientists are concerned about the extinction of insular species. Finally, links with local research institutions will be strengthened, as well as with other institutions devoted with Macaronesian flora, namely with Madeira and Azores Universities.

VISUALISING THE DEVELOPMENT OF EVOLUTIONARY THOUGHT: A GRAPHICAL OUTREACH PROJECT

Tania Jenkins¹, Miriam Quick², Stefanie Posavec¹ ¹Christe, Department of Ecology and Evolution, University of Lausanne, Switzerland ²freelance researcher, United Kingdom ³freelance designer, United Kingdom tania.jenkins@unil.ch

The general public remains confused about evolution. To prevent creationism from taking hold, there is a pressing need for both clarification and – just as importantly – inspiration. The aim of this infographic is to trace the progression of evolutionary thought from pre-Darwinian times to the present in a visually striking way. Standing at the intersection of science and art, this work shows through an innovative use of colour, text and graphics how evolution by natural selection has come to be established as the dominant force explaining the diversity of life. Our aim is to communicate the facts of evolution – but also inspire and excite – using a creative, compelling design to attract the eyes of people not normally interested in science. This poster will be made available both on request and online.

THE MISMATCH BETWEEN EDUCATION AND RESEARCH ON EVOLUTIONARY BIOLOGY IN SPAIN

Juan Arroyo¹, Manuel Serra², Miguel Verdú¹, Fernando González-Candelas² ¹Department Plant Biology and Ecology, University of Seville, Spain ²Inst de Biodiversidad y Biología Evolutiva, University of Valencia, Spain ³CIDE, CSIC, Spain

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Since the Renaissance, cultural traditions, moral prejudices and social structure have hindered the development of modern science in Spain, with a minor burst during the 18th c. Political instability during second half of the 19th c. was also coincident with the development of Darwinian ideas, which had little impact on the Spanish cultivated class, with some noteworthy exceptions. This situation spanned most of the 20th c. until the economical burst of the 60's, when science became a pursuit of increasing interest for Spanish leaders. With the advent of democracy in the late 70's and early 80's scientific achievements became a prominent goal progressively assumed by a rapidly growing middle class and normalized research programs and procedures were established. As a result, Spain currently ranks ninth in the world in the amount of relevant scientific production. Biology has shown a paradigmatic progress, and evolutionary biology has been one of its fastest growing fields. In contrast, teaching of evolution at different educational levels, from secondary schools to postgraduate programs, has been surprisingly neglected. Thus, evolutionary biology shows a mismatch between teaching ad research that deserves an explanation. Here we present statistics that document the extent of the mismatch. By comparing with other countries and other science fields, we examine what the possible causes might be. We propose that evolutionary-like processes such as founder effects and drift due to initial low number of practitioners, or selective processes against individuals in non-organized groups, might explain this distortion. Mirroring evolutionary processes, social processes such as those involving a scientific community have some delay in showing the consequences, which we aim to foresee in the hope of preventing them.

Symposium

30. Phylogenetics and Phylogeography

21, 22, 23 and 24 August



Program

Wednesday 21 August

Session(s): 7, 8

Thursday 22 August

Session(s): 9

Friday 23 August

Session(s): 10, 11, 12, 13

Saturday 24 August

Session(s): 14

Organisers: Thomas Schmitt and Richard Nichols

Invited speakers: Lacey Knowles and Emily Lemmon

Description:

This is a wide scope session mostly addressing aspects in the field of geographical distribution pattern of genetic information. Novel data enlarging our knowledge on range dynamics of species are highly welcome as well as presentations advancing the analytical tool and methodological aspects in this field. However, this session not only includes phylogenetics and phylogeography approaches, but also landscape genetics and a wide range of topics on the evolutionary processes across species ranges and of conservation aspects in larger scale population genetics.

D24SY30IT10:30R1

TESTING THE WHEN, WHERE, AND HOW OF DIVERGENCE WITH SPECIES-SPECIFIC PREDICTIONS OF GENETIC VARIATION UNDER ALTERNATIVE HYPOTHESES

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Recent developments at the molecular level, as well as computational advances that include the forging of diverse approaches, have the potential to dramatically increase the insights of phylogeographic studies. I will describe one such approach - namely, testing hypotheses based on species-specific predictions about patterns of genetic variation under different scenarios. By example, I will show how the coupling of species distributional models with demographic and genetic models can be used to test alternative hypotheses in an endemic lizard from southwest Australia, Lerista lineopunctulata, about the role of geographic, environmental, and changing climatic conditions in structuring genetic variation across the landscape. Because the lizard is distributed along the coastal sand plain or dunes, a habitat that is discontinuous today and that has shifted over time, it is conceivable that population genetic divergence could reflect the contemporary habitat configuration, which limits migration among the small geographically isolated populations, or colonization processes associated with historical shifts in the species distribution, given that a habitat specialist would track climate-induced habitat shifts. With the modeling of these alternative scenarios and tests of multilocus data using approximate Bayesian computation, I show that the dynamic history of shifting environmental conditions since the last glacial maximum, not the geographic configuration or populations or the contemporary environment itself, is the primary factor structuring genetic variation. I also highlight how this approach can be expanded to multiple species, where the goal is not to quantify concordance, as with traditional comparative phylogeographic approaches, but instead to understand why patterns of genetic variation are (or are not) similar across species.

D24SY30IT11:18R1

ANCHORED PHYLOGENOMICS: ACCELERATING THE RESOLUTION OF LIFE

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The field of phylogenomics is undergoing a revolution, enabled by new methods of data collection that leverage both genomic resources and recent advances in DNA sequencing. We developed a costefficient and rapid approach to obtaining data from 100s to 1000s of loci and individuals for deep and shallow phylogenetic studies. Specifically, we designed probes for target hybrid enrichment of 100s of loci in conserved anchor regions of genomes (flanked by less conserved regions). We enriched genomic DNA libraries for these anchor regions, and sequenced these targets using high-throughput sequencing. The resulting data sets contain 100s of loci with low levels of missing data and high levels of phylogenetic information across taxonomic scales and produce phylogenies with high levels of resolution. This approach is expediting resolution of deep-scale portions of the Tree of Life and greatly accelerating resolution of the large number of shallow clades that remain unresolved. The combination of low cost (~1% of the cost of traditional Sanger sequencing and ~3.5% of the cost of highthroughput amplicon sequencing for projects on the scale of 500 loci × 100 individuals) and rapid data collection (\sim 2 weeks of laboratory time) make this approach tractable even for researchers working on systems with limited genomic resources. Here, I present new work from vertebrates and unpublished data from various non-vertebrate (e.g., Coleoptera, Hymenoptera, etc.) and plant (e.g., Angiosperm) clades; I also discuss future directions and new applications.

30. Phylogenetics and Phylogeography

D21SY30RT15:45R2

PHYLOGENETIC PATTERNS OF DIVERSIFICATION AND COMMUNITY STRUCTURE REVEAL THE ROLE OF ECOLOGICAL FACTORS IN GENERATING AND SHAPING NEOTROPICAL MIMETIC BUTTERFLY DIVERSITY

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The Neotropic is the most diverse region in the world. Identifying the causes underlying the observed diversity and spatial structure and unraveling how different factors interact remain outstanding questions. Addressing these questions in the Neotropics provides a unique opportunity to understand the processes that shape the structure of ecosystems. Recent advances in molecular phylogenetics, such as novel tests for diversification scenarios or community ecology phylogenetics, provide new insights into biodiversity questions. The Ithomiini tribe is a species-rich group of Neotropical butterflies, which consists of ca. 380 species widely distributed from the lowlands to 3000 m in the Andes. All species are involved in Müllerian mimicry, whereby different species protected by chemical defenses converge to share the same warning color patterns (mutualistic interactions). Here we combine phylogenetic, trait and species assemblage (community) data to investigate the effects of color patterns and adaptation to altitude in generating and shaping the diversity of Ithomiini.Comprehensive phylogenetic and trait analyses of several genera show that shifts in altitude and color patterns are both involved in diversification. In parallel, phylogenetic analyses of 15 communities along an altitudinal gradient show that both adaptation to altitude and mimetic interactions act as ecological filters and contribute in shaping species assemblages. Moreover, we find that both factors are not independent, and species that share color patterns also share altitudinal niche more often than predicted by phylogeny alone. The two approaches undertaken here are complementary and provide support for a similar scenario, whereby ecological factors, especially positive interactions, have been instrumental in generating and shaping biodiversity. Taking other types of positive interactions into consideration may shed light on why some groups have diversified dramatically, and why others have not.

30. Phylogenetics and Phylogeography

D21SY30RT16:09R2

LARGE BODY SIZE AS A KEY INNOVATION FACILITATING RAPID RADIATION AND GEOGRAPHICAL EXPANSION IN MALAGASY DUNG BEETLES

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Dung beetles form highly competitive communities and therefore are likely to show patterns of diversification consistent with the diversity-dependent model of radiation, whereby the progressive filling of ecological space, characterized by an initial rapid accumulation of lineages, is followed by a slowdown in net diversification rate. In such cases, further diversification will only be possible following a "key innovation", which would decouple the rate of diversification in the "innovative" lineage from that in the main radiation. Here, we integrate a well-represented species-level dated phylogeny of the most recent but large dung beetle radiation in Madagascar (Nanos-Apotolamprus radiation, Canthonini) with data on species' geographical ranges, abundances and body sizes. Results demonstrate an overall decrease of lineage accumulation through time. However, we observe a significant secondary burst of speciation associated with one clade composed of large-bodied species (Clade L, for large), consistent with the "key innovation" model. The secondary increase in diversification rate is associated with a significant increase in body size and striking geographical expansions. It is common in dung beetle communities for large species to be competitively superior to small ones in exploitative competition and hence we suggest that a significant increase in body size has allowed the large species in Clade L to diversify independently of the species in the rest of the lineage. Furthermore, we show that the increased speciation rate in Clade L is most probably associated with competitive release. This is supported by the observation that the total biomass of many species in Clade L is far greater than the total biomass in any of the remaining species. Finally, species in this successful clade have small but almost entirely allopatric distributions: the increase in species number is hence importantly due to divergence in their geographical occurrences.

30. Phylogenetics and Phylogeography

D21SY30RT16:33R2

COMPARATIVE PHYLOGEOGRAPHY OF AFRICAN RAIN FOREST TREES: A REVIEW OF GENETIC SIGNATURES OF VEGETATION HISTORY IN THE GUINEO-CONGOLIAN REGION

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The biogeographic history of the African rain forests has been contentious. Phylogeography, the study of the geographic distribution of genetic lineages within species, can highlight the signatures of historical events affecting the demography and distribution of species (population fragmentation or size changes, range expansion/contraction) and, thereby, the ecosystems they belong to. The accumulation of recent data for African rain forest plants now enables a first biogeographic synthesis. In this review, we explain which phylogeographic patterns are expected under different scenarios of past demographic changes; and we give an overview of the patterns detected in African rain forest trees to discuss whether they support alternative hypotheses regarding the history of the African rain forest cover. Phylogeographic patterns indicate that most of the widespread species showing nowadays a near continuous spatial distribution have been fragmented in the past. The major genetic discontinuities in the region support the role of refugia during climatic oscillations, and at a large scale they often match the main phytogeographic subdivision of the Guineo-Congolian region. However, patterns vary substantially among species and they provide only partial support for current hypotheses on the location of Pleistocene forest refuges. Among the patterns shared by many species, we highlight a recurrent genetic discontinuity between the north and the south of the Lower Guinean region which matches the climatic hinge on each side of which dry and wet seasons are inverted. Whether this discontinuity results from past forest fragmentation or current climate conditions is discussed.

D21SY30RT16:57R2

THE EFFECT OF HABITAT ON PLANT DIVERSIFICATION: A MACROEVOLUTIONARY APPROACH

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The close relationship between plants and their habitats is a research subject of long-standing interest. However, the macroevolutionary patterns of this association are still unclear. For instance, it is not known whether habitat conditions affect the diversification of plant lineages. Moreover, few studies have investigated if ecological conservatism is a general phenomenon with comparable patterns across habitats and lineages. Here we present an analysis of habitat evolution in Saxifragales, an angiosperm clade exhibiting diverse morphologies and habitats. Using well-resolved phylogenies built with a super-matrix approach and habitat data for ~1000 taxa (50% of the group) we investigate a) The pattern of habitat evolution: the most likely ancestral habitat and the rates of transition across the different classes of habitats b) The association between diversification rates and habitat, determining if the speciousness of certain lineages is related to their habitat; or, if on the contrary, certain habitats are linked to higher extinction rates c) Ecological conservatism across lineages and habitats: if the adaptation to certain habitats constrains diversification into other environments, and whether the constrains are equivalent for different habitats Our results show that from a forest-inhabiting ancestor, the Saxifragales have transitioned into a variety of environments. Some of these habitat shifts appear to have triggered bursts of diversification. Seemingly harsh environments, such as rock cliffs and the tundra, are linked to high diversity and have been occupied and abandoned repeatedly throughout evolution. Conversely, deserts contain only ancient, species-poor lineages, and the transition into an aquatic medium seems irreversible. This indicates that although adaptation to extreme conditions can lead to evolutionary cul-de-sacs, this phenomenon is complex and very dependent on as-yet unknown underlying physiological and morphological changes.

D21SY30RT17:45R2

FUNCTIONAL HAEMOGLOBIN DIVERGENCE BETWEEN BANK VOLE POPULATIONS OF DIFFERENT REFUGIAL ORIGINS

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Much has been learned about the post-glacial partitioning of phylogeographic lineages in various species, yet little is known of the extent to which their distributions have been determined by adaptive differences. We have addressed this issue by studying the genetic basis of haemoglobin (Hb) polymorphism in the bank vole (Clethrionomys glareolus). It is evident that at the end of the last glaciation, bank voles colonized Britain from two different source populations that were already isolated and genetically divergent. Regions colonised by the two lineages can be identified from mtDNA phylogeography and are fixed for different electrophoretically detectable Hb variants. Since modifications of Hb structure and function often play a key role in adaptation to climate and altitude, it has been suggested that the geographical pattern of bank vole Hb variation in Britain may reflect some type of environmental selection favouring the HbS type in the north and the HbF type in the south. We have cloned the genes coding for bank vole Hb by RACE-PCR and assessed their relative expression by RNA-Seq, in order to determine whether the alpha or beta chain polymorphism, and how many loci, underlie the two Hb variants. The results demonstrate that a single amino acid replacement mutation, 52(D3)Ser/Cys, in the major expressed beta globin gene underlies the difference between HbS and HbF. The same polymorphism segregates also at the second, minor expressed beta globin gene, suggesting a possible functional interplay between the two paralogs. The exposed location of the thiol group of 52(D3)Cys on the surface of HbF molecule and the ability to form intermolecular disulphide bonds are indicative of its reactivity and therefore potential involvement in physiologically relevant redox reactions. We suggest that antioxidant potential of the reactive Cys in HbF may convey selective advantage over HbS and contribute to the geographical partitioning of the two bank vole lineages in Britain.

D21SY30RT18:09R2

COMPARATIVE PHYLOGEOGRAPHY OF THREE TREE SPECIES FROM THE ALBANY SUBTROPICAL THICKET OF SOUTH AFRICA: TESTING CORNERSTONE HYPOTHESES OF A LARGE-SCALE CONSERVATION NETWORK

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An ambitious large-scale network of conservation areas and corridors has been designed with the aim of preserving the evolutionary patterns and processes of the Albany Subtropical Thicket biome of South Africa. Two main hypotheses, largely based on expert opinion, formed the foundation for this conservation plan: 1) drainage basins are discrete biogeographical, and thus evolutionarily-distinct, units, and 2) long-distance movement of the primary agents of disturbance in this landscape, African elephants (Loxodonta africana), is critical for ecosystem functioning. Here we test these hypotheses using chloroplast phylogeography of three tree species with similar ecologies but different seed dispersal syndromes, specifically Nymania capensis (Meliaceae; inflated wind-dispersed seeds), Pappea capensis (Sapindaceae; small bird-dispersed seeds) and Schotia afra (Fabaceae, large seed-pods dispersed by elephants and other large mammals). Each drainage basin was found to have isolated populations of the wind- and bird-dispersed species, whereas the elephant-dispersed species has a single regional meta-population that extends across all the drainage basins. These results suggest that the deeply-incised drainage basins along the coastal lowlands of South Africa are unique phylogeographic, as well as biogeographic, units for species that that lack long-distance dispersal mechanisms over such a topographically-complex landscape. At present, elephants and many other large herbivores are restricted to an extremely reduced and fragmented proportion of their historical distribution along the coastal lowlands. The phylogeographic patterns of S. afra highlight the importance of re-establishing the free movement of megaherbivores across the regional landscape for this and other elephant-dispersed plants. These phylogeographic patterns offer pivotal support for the proposed network of conservation corridors for this biome.

D21SY30RT18:33R2

AVIAN MALARIA DIVERSITY IN SOUTHERN MELANESIA: EXPLORING POTENTIAL INFLUENCES ON DISTRIBUTION PATTERNS AND DIVERGENCE OF HOST BIRD SPECIES

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Host-pathogen interactions have the potential to influence broad scale ecological and evolutionary processes in host populations, influencing levels of endemism, divergence patterns and distributions. The development of localized co-adapted host-pathogen relationships could drive divergent selection and limit gene flow among populations of the same species or limit range expansions of closely related species. To examine the potential for pathogens to direct these types of processes in natural host populations, we explored the phylogenetic diversity and prevalence of avian malaria parasites, and the distribution of lineages across islands and host species, in southern Melanesian bird communities. We uncovered high avian malaria diversity, including many rare lineages, pointing to high estimates of undiscovered lineage richness in the region. While significant phylogeographic structuring of parasite lineages was detected, most genetic variation was distributed within rather than between island locations. Only a small number of lineages could be considered generalist with respect to host species infection. Parasite lineage-host associations were evident at the host family and host species level, and in a few cases, at the local host population level. Prevalence varied significantly among islands, but not among host species, and for three widely sampled host species, there was a significant interaction term demonstrating variation in prevalence in different populations of the same species. The heterogeneity demonstrated in both the complement and the prevalence of parasite lineages infecting local avian communities likely exposes species to a mosaic of disease-related selection pressures across their naturally fragmented distributions in southern Melanesia. Therefore interactions between avian malaria parasites and their avian hosts are an important factor to consider when examining avian divergence and distribution patterns in this region.

D21SY30RT18:57R2

PHYLOGEOGRAPHY AND MORPHOLOGY OF THE SOUTH AMERICAN ANNUAL KILLIFISH GENUS, AUSTROLEBIAS

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The genus Austrolebias consists of over 30 species of freshwater annual killifish. These fish reside in ephemeral pools across a wide region of South America including parts of Brazil, Uruguay, Paraguay, Argentina and Bolivia. Austrolebias possess a peculiar and interesting life-history. Males and females dive together into the muddy substrate of the pond and deposit their gametes. Later, these adults are often seen dying in shallow water as the ponds completely evaporate during the beginning of the dry season. The eggs then go through several stages of diapause until hatching is triggered by the earliest wet season rains, after which the fish grow rapidly and the process is repeated. The large geographic range of Austrolebias means that the genus is subject to considerable variation in climate. I reveal which climate variables are most important in determining the distribution of Austrolebias by using data collated from my own collection trips, primary literature, online biodiversity databases as well as the records of amateur collectors. Larger species of Austrolebias can be up to 13cm in length, the smallest only 4cm. Recent phylogenetic work has shown that large species have been derived from small species in at least 3 separate instances. I use phylogenetic independent contrasts to investigate whether any bioclimatic variables can explain this variation in size. In addition, species distribution models (SDMs) are built using Maxent and subsequently compared using ENMTools in order to identify whether those species that are morphologically similar possess a similar climatic niche. The nature of the Austrolebias life cycle suggests that it is sensitive to climate change. Current SDMs are compared to 2050 SDMs using the HadCM3 model with multiple emission scenarios to predict the future range shifts of *Austrolebias* in order to discern the effect climate change will have on this genus.

D22SY30RT10:30R9

THE HIDDEN HISTORY OF THE SNOWSHOE HARE (LEPUS AMERICANUS): DEEP INTRASPECIFIC DIVERGENCE AND MASSIVE MITOCHONDRIAL DNA INTROGRESSION

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The analysis of molecular variation in wild populations often allows uncovering fundamental evolutionary processes that could otherwise remain unknown. Lepus americanus is the most widespread North American hare and its ecology has been vastly studied. However, knowledge on its evolutionary history is very scarce. A recent work based on microsatellite and mtDNA data suggested that this species is structured in three major units, which may have resulted from fragmentation during Pleistocene glaciations. Also, it suggested that the mtDNA lineage of one of these units is more closely related to that of a neighboring southern species, Lepus californicus, which could result from introgression or sharing ancestral polymorphism. Here, we sequenced nine loci, representative of all inheritance compartments (6774 bp), and shed light on the evolutionary history of *L. americanus* (48 specimens), taking also into account the evolution of L. californicus (30 specimens) and a third North American species, L. townsendii (18 specimens). Using a coalescent-based species tree inference method we show that the three evolutionary units of L. americanus have genealogical relevance and that one of the groups, widely distributed along the Boreal region, diverged at about 2 Mya, i.e. at the same time scale of the split between L. californicus and L. townsendii (1.6 Mya), without obvious morphological or ecological divergence. Also, coalescent simulations based on parameters of speciation inferred using the IM model, suggest that massive ancient mtDNA introgression occurred 680 kya from L. californicus into the Pacific Northwest populations of L. americanus. Interestingly, the specimens in this region have a peculiar morphology, since some do not molt to the white winter coat, an uncommon phenotypic variation in *L. americanus*. The snowshoe hare is thus a valuable model to understand cryptic divergence, adaptation and speciation in the presence of gene flow.

D22SY30RT10:54R9

INFLUENCE OF SEASCAPE AND MIGRATORY PATTERNS ON COASTAL BROWN TROUT GENETIC STRUCTURE (SALMO TRUTTA)

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Understanding how anthropogenic and natural factors influence movement patterns of wild organisms is of prime importance in conservation biology. Most animal species live in spatially and temporally heterogeneous landscapes where resources are unequally spread. Consequently, individuals must adjust their migratory behaviour to avoid adverse environmental conditions and reach suitable areas for feeding and breeding. In the present work, we studied populations of brown trout (Salmo trutta) from the rivers of the French Channel coast that display a large continuum of migratory strategies from complete freshwater residence to anadromy (sea trout). To clarify the influence of life-history strategies and natural environmental factors on population connectivity, we used an integrated approach that combines GIS data on landscape structure, patterns of life history variation and inferences of dispersal based on neutral genetic markers. Landscape genetic analyses revealed a hierarchical genetic structure with two main genetic units corresponding to the eastern and western marine ecoregions of the English Channel delineated by the Cotentin peninsula. Within these two clusters, the spatial variation of migratory strategies appears to influence the patterns of isolation by distance and we observed a latitudinal increase in population genetic isolation. Moreover, least-cost path modelling suggests that sea trout preferentially disperse through soft bottom areas in brackish and shallow waters where they find their favourite prey. Overall, these results point out the need to take into account the spatial variability of life history and landscape heterogeneity at large scale when studying the ecology and evolution of long distance migratory animals.

D22SY30RT11:18R9

POLLINATOR-RESTRICTIVE FLOWER TRAITS DETERMINE EVOLUTIONARY OPTIMA WITH DIFFERENTIAL DIVERSIFICATION RATES IN BIFID TOADFLAXES (*LINARIA* SECT. *VERSICOLORES*)

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The role of flower specialization in plant speciation and evolution remains controversial. Here we analyzed the evolution of flower traits restricting the access to pollinators in the bifid toadflaxes (*Linaria* sect. *Versicolores*), a monophyletic group of c. 30 species and subspecies with highly specialized corollas. A time-calibrated phylogeny based on both nuclear and plastid DNA sequences was obtained using a coalescent-based method, and flower morphology was characterized by means of morphometric analyses. Using recently-developed methods, directional trends in morphological traits and trait-dependent diversification rates were jointly analyzed, and morphological shifts were reconstructed along the phylogeny. Our results indicated that a restrictive character state (narrow corolla tube) may be ancestral to Linaria sect. Versicolores. After its early loss in the most species-rich clade, this character state has been convergently reacquired in multiple lineages of this clade in recent times, yet it has exerted a negative influence on diversification rates. Comparative analyses and pollinator surveys suggest that the narrow- and broad-tubed flowers are evolutionary optima representing divergent strategies of pollen placement on nectar-feeding insects. We therefore suggest that opposing individual-level and species-level selection pressures may have driven the evolution of pollinator-restrictive traits in bifid toadflaxes.

D22SY30RT11:42R9

CO-DIVERGENCE BETWEEN BACTERIA AND HOST: CONSEQUENCES OF WOLBACHIA INFECTION IN THE CHORTHIPPUS PARALLELUS HYBRID ZONE

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Divergence between two subspecies of the grasshopper Chorthippus parallelus has been extensively studied for evolutionary purposes. They form a hybrid zone in the Pyrenees between the Iberian endemic subspecies C. p. erythropus and the continental European subspecies C. p. parallelus (1). Wolbachia infection in pure and hybrid individuals of these subspecies has been characterized by PCR analysis of their 16S rRNA sequences and a Multi-Locus Strain Typing System (MLST) (2). Phylogenetic and phylogeographic studies based on previous host phylogeny, as well as this MLST approach, show that Wolbachia co-diverges with its host. In addition, extrapolation of the divergence time between bacterial strains allows us to propose than Wolbachia infected C. parallelus during host speciation. This fact, joint to the cytoplasmic incompatibility and other effects induced by this bacterium in natural populations of C. parallelus (3, 4) support that co-divergence between bacteria and host contributes to explain the current situation of the hybrid zone. References (1) Shuker DM, et al. 2005. "The genetic basis of speciation in a grasshopper hybrid zone". En "Insect Evolutionary Ecology". CABI Publishing, Oxford University Press, pp 427-454. ISBN 0851998127. (2) Baldo L, et al. 2006. Multilocus Sequence Typing System for the Endosymbiont Wolbachia pipientis. Appl Environ Microbiol 72:7098-7110. (3) Bella, J.L, Martínez-Rodríguez, P., Arroyo-Yebras, F., Bernal, A., Sarasa, J., Fernández-Calvín, B, Mason, P.L. & Zabal-Aguirre, M. 2010. "Wolbachia infection in the Chorthippus parallelus hybrid zone: evidence for its role as a reproductive barrier". Journal of Orthoptera Research, 19 (2): 205-212. (4) Sarasa, J., Bernal, A., Fernández-Calvín, B. & Bella, J.L. 2013. Wolbachia induced cytogenetical effects, as evidenced in Chorthippus parallelus (Orthoptera). Cytogenetic and Genome Research 139: 36–43.

D23SY30RT10:30R8

DISPERSAL, GENE FLOW AND ECOLOGICAL SPECIATION IN LAMPREYS

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In species with complex life histories estimates of dispersal rates can be obtained from genetic data. This approach allows comparing levels of intraspecific gene flow among species with different dispersal abilities. Lampreys evolved as either anadromous species (i.e. reproducing in freshwater but growing mainly at sea) with a parasitic lifestyle at sea or as non-parasitic freshwater resident species. Anadromous species home to their natal river or disperse to other rivers and should thus display higher level of gene flow among populations than resident species. The three European lamprey species allow testing this prediction: Lampetra planeri (LP) is resident in freshwater while L. fluviatilis (LF) and *Petromyzon marinus* (PM) are anadromous. LF and LP are morphologically and phylogenetically very similar and they may represent two ecotypes of a single species. We investigated the genetic structure among LP, LF and PM populations in France to i) test whether levels of gene flow are higher in anadromous than in resident species and ii) measure the level of gene flow among sympatric LF and LP populations. We used microsatellite loci to genotype 665 LF and LP and 380 PM individuals. Our results show a gradient of increasing genetic differentiation among lamprey species (PM < LF < LP) suggesting that populations of anadromous species are connected by higher levels of gene flow than LP populations. Interestingly, we detected some gene flow among sympatric LF and LP populations supporting the hypothesis that these species are actually two ecotypes of a single species possibly at an early stage of ecological speciation. In addition, estimates of genetic diversity were significantly higher in LF than LP populations. These results have important implications for conservation management through the definition of evolutionary significant units and species boundaries. They also suggest that the connectivity between putative LP and LF ecotypes should be maintained.

D23SY30RT10:54R8

GENETIC BASIS AND GEOGRAPHICAL PATTERNS OF ADAPTIVE DIVERGENCE IN AN INTERTIDAL SNAIL

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At the onset of ecological speciation with gene flow, an ancestral species splits into ecotypes with incomplete reproductive isolation. In multiple systems, ecotypes coexist in many geographical locations, raising the question whether the genetic basis of divergence is identical across the whole range. While some loci may be under divergent selection on large geographical scales (reflecting a spread of favourable alleles), others might be involved in divergence only locally (potentially causing "parallel evolution"). Geographically close locations with a shared colonization history may be expected to share more divergently selected loci than distant ones. Our project aims at estimating the relative contribution of globally and locally selected alleles to divergence between ecotypes of the marine snail Littorina saxatilis. Using snails from Spain, Sweden and the United Kingdom, we performed an RNA-sequencing experiment and analysed the resulting allele frequency data. Because Swedish and British locations probably have a shared postglacial colonization history, we expected them to share loci under divergent selection. We found a large number of single nucleotide polymorphisms (SNPs) which showed differentiation between ecotypes only locally, indicating parallel evolution. In Spain there were more SNPs with significant differentiation between ecotypes than in Sweden and the UK, potentially reflecting stronger divergence between ecotypes. While transcriptomewide differentiation between Swedish and UK samples was lower than their differentiation from Spain, we did not find a higher number of shared loci under selection. However, we identified >1000 SNPs showing increased differentiation between ecotypes across all three countries, suggesting a shared origin of divergence. Our results contribute to the understanding of the genome as a mosaic of loci with partly independent evolutionary histories and demonstrate how genomic patterns of selection vary across space.

D23SY30RT11:18R8

TRACING THE ORIGINS OF DOMESTICATION IN THE WINE YEAST SACCHAROMYCES CEREVISIAE

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The domestication of plants and animals represents a milestone in human history because it triggered critical transitions of human civilizations. Recent studies suggest that the yeast Saccharomyces cerevisiae, which is used nowadays in the fermentation of wine and other alcoholic beverages worldwide, illustrates the case of a microbe that has been also domesticated. However, the vast wealth of knowledge that has been gathered for the understanding of crop and livestock domestication contrasts markedly with our ignorance on microbe domestication. Recent advances in sequencing technology are providing novel opportunities for microbe population and evolutionary studies through the availability of genome-wide molecular markers that allow an unprecedented fine scale resolution of population dynamics and organismic evolution. Here, we have surveyed sequence variation using whole-genome sequencing in a dataset of S. cerevisiae strains that were isolated from natural environments (putative wild populations) and human-associated niches (putative domesticated populations), from different geographic regions at a global scale. The hypothesis to be tested is that the wild ancestors of wine yeasts are found close the main center of origin and spread of winemaking. We found a distinct and previously undescribed phylogenetic lineage of S. cerevisiae that is the closest known wild relative to the wine yeast group and is distributed along the Mediterranean basin. We propose that these two lineages diverged only recently, a view that is reinforced by the proportion of shared alleles and the low number of fixed differences between them. We discuss the significance of an excess of rare variants in the wine group relative to wild strains. The work described here seeks to provide new insights into the history of domestication and improve our knowledge on the ecology of this important microbe.

D23SY30RT11:42R8

FROM AFRICA AND BACK AGAIN: BIOGEOGRAPHY AND DIFFERENTIATION OF A SPECIALIST WEST-MEDITERRANEAN BUTTERFLY IN A FRAGMENTED GLACIAL REFUGE

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The Mediterranean area is well known for retaining and generating biodiversity during climate and habitat shifts taking place since the end of the Miocene. This important biodiversity hotspot is both the source and the result of a complex process of differentiation and interaction between evolving lineages and their changing environment. However, regarding stationary Mediterranean specialists, which are expected to benefit and expand northwards with climate amelioration, little is still known. Using the butterfly *Euchloe tagis*, an Atlanto-Mediterranean ecological specialist with a fragmented and restricted distribution, this study attempts at answering pertinent biogeographic aspects on the species and its interaction with the changing ecosystem. We sequenced a 599bp barcode region fragment of the COI gene from all major populations to better understand current its genetic patterns and evolutionary history. Main results suggest an origin in North Africa, from where the sister species *Euchloe pechi* is endemic. While one of the Moroccan populations diverged earlier from the remaining stock, all remaining are more closely related despite the existence of differentiated major mitochondrial lineages. It seems that Iberia has played a pivotal role in the generation of diversity in this species: two consistent genetic clusters are partly sympatric whereas French and Italian populations are considerably different. The second Moroccan population from the Rif Mountains shows little segregation from French samples and this issue is discussed on a phylogeographic framework supported by molecularclock analysis. This pattern suggests this species experienced polycentric refugia during climate cycles, leading to such genetic structure and a putative re-colonization of Africa. Moreover, the neutral genetic pattern reported in this study challenges current taxonomy based on morphological differentiation and suggests a more complex evolution of this species.

D23SY30RT14:00R8

MAKING THE BEST OF YOUR 454 DATASET USING THE 4PIPE4 ANALYSIS PIPELINE – FROM AN UNKNOWN GENOME TO A DETAILED SNPS ANALYSIS: A CASE STUDY WITH A QUERCUS SUBER DATASET

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The widespread use of Next Generation Sequencing (NGS) techniques and its application to non model organisms provided biologists with large amounts of genetic data and the ability to address problems which were untreatable just a few years ago. However, the frequent lack of reference genomes on nonmodel organisms usually creates difficulties to researchers when trying to answer specific genetic questions, such as finding candidate genes or looking for intra and inter-population variation. For instance, mining SNPs in NGS datasets of anonymous pooled individuals that cannot be compared to a reference is still not a simple task. Here is described 4Pipe4, a NGS data analysis pipeline, optimized for SNP mining in the aforementioned datasets, particularly on Roche 454 data. In order to assess its efficiency, a dataset of anonymous pooled individuals of Quercus suber (Cork Oak), which does not have a reference genome available, was analysed with 4Pipe4, and a subset of tenths of SNPs detected by the pipeline was randomly selected and sequenced in an array for validation. The results of the genotyping array were explored to: a) provide insights on population structure and gene flow patterns and b) make an association study with environmental factors such as temperature, precipitation or drought periods. This combined approach of 454/genotyping array with the 4Pipe4 pipeline proved to be a very efficient and cost effective way to obtain validated and mapped SNPs from orthologous regions, for population genomics studies.

D23SY30RT14:24R8

IGNORING HETEROZYGOSITY BIASES PHYLOGENOMIC ESTIMATES OF DIVERGENCE TIMES: IMPLICATIONS FOR THE EVOLUTIONARY HISTORY OF *MICROTUS* VOLES

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Challenges for the phylogenetic reconstruction of the evolutionary history of closely related organisms may arise from the presence of unsorted lineages and a relatively high proportion of heterozygous sites that are often difficult to incorporate in the analysis. Here we performed a phylogenomic reconstruction of the evolutionary history of the common vole (*Microtus arvalis*) in Europe with a special focus on the influence of heterozygosity on the estimation of intra-specific divergence. We provide a novel strategy to integrate heterozygous information in existing phylogenetic programs by repeated random haplotype sampling (RRHS) from sequences with multiple unphased heterozygous sites. We evaluate the impact of full, partial or excluded information from heterozygous sites in tree reconstructions on divergence time estimates. All results consistently show four deep and strongly supported evolutionary lineages in our genomic vole sequence data. These lineages diverged at the end of or shortly after the last glacial maximum based on a calibration using radiocarbon-dated archaeological information. However, the incorporation of information from heterozygous sites had a significant impact on absolute and relative branch length estimations. Ignoring heterozygous information led to overestimation of divergence times between the evolutionary lineages of *M. arvalis*. Computer simulations confirmed the high power of our empirical data set to recover the evolutionary history of the species and allowed to examine potential sources of bias in different analytical approaches. In general, the exclusion of heterozygous sites from evolutionary analysis may cause biased and misleading divergence time estimates particularly in closely related taxa.

D23SY30RT14:48R8

THE MAINTENANCE OF MITOCHONDRIAL GENETIC VARIATION BY NEGATIVE FREQUENCY-DEPENDENT SELECTION

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The processes that maintain biological and genetic diversity are not fully understood. Mitochondrial genes, which encode proteins involved in a key energetic pathway in eukaryotic cells, often show high levels of standing genetic variation. This observation is especially puzzling, given the accumulating evidence for life history and fitness effects of mitochondrial genetic variation, because selection should rapidly exhaust genetic variation in the mitochondrial genome. Negative frequency-dependent selection, where the relative fitness of a genotype is inversely related to its frequency in a population, provides a potent and potentially general process that can maintain mitochondrial genetic polymorphism. However, empirical tests of this possibility are lacking. Here, we present experimental evidence that negative frequency-dependent selection acts to maintain polymorphism in mitochondrial genes. We assessed the change in mitochondrial haplotype frequencies over 10 generations of experimental evolution in a large number of seed beetle populations, where haplotypes competed for propagation to subsequent generations. We found that haplotypes consistently increased in frequency when they were initially rare and decreased in frequency when initially common. Furthermore, the strength of frequency-dependent selection was contingent upon epistatic mitonuclear interactions, directly supporting the tenet that intergenomic epistasis is important in mtDNA evolution. Our results have important implications for the use of mtDNA haplotype frequency data to estimate population level phenomena and they revive the general hypothesis that negative frequency-dependent selection may commonly facilitate genetic polymorphism in life history traits.

D23SY30RT15:12R8

HOW TO USE RADSEQ DATA TO INFER PAST DEMOGRAPHY IN A MULTI-LOCUS COALESCENT-BASED FRAMEWORK

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Next-generation sequencing (NGS) methods that allow for genome scan in multiple individuals are expected to trigger a new golden age in phylogeography, phylogenetics and population genetics likely merging these fields with ecological genomics. Neutral and adaptive processes are now likely to be addressed with the same toolkit of molecular markers. However, the implementation of whole-genome data in ecological adaptations studies is far advanced than in the investigation of neutral evolutionary dynamics. This is mainly due to the most common output data type of most NGS methods (e.g. unlinked SNPs) in contrast with the historical importance of using gene trees, mainly through coalescent-based analytical approaches, in phylogeography and phylogenetics. Nevertheless, solutions that can make the set of excellent statistical tools for DNA sequences analysis, employed in phylogeography and phylogenetics so far, available to deal with NGS data are of the utmost importance. Here we will show how RAD sequencing data can be used to address classical questions in phylogeography and we will also propose a novel method to use RADseq data to infer past demography in a coalescent-based framework. Two applications of the proposed approach will be shown: i) the analysis the past population history of a king penguin (*Aptenodytes patagonicus*) colony breeding on Crozet archipelago and ii) the study of the past evolutionary dynamics of two populations of African porcupines (Hystrix cristata and H. africaeaustralis).

D23SY30RT15:45R8

ASSESSING THE CONFOUNDING EFFECT OF POPULATION STRUCTURE ON BAYESIAN SKYLINE PLOT INFERENCES OF DEMOGRAPHIC HISTORY

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It is well known that population structure can confound coalescent-based inferences of past population size changes when it is not properly accounted for. Although the vast majority of species are genetically structured to some extent, few studies have quantified how different types and levels of structure might confound demographic inference. Among the most widely applied demographic inference methods is the Bayesian skyline plot (BSP and the derived EBSP), which assume a panmictic model without structure. We simulated DNA sequence data under a variety of scenarios involving structured populations with variable levels of gene flow and analysed them using EBSPs as implemented in the software package BEAST. Results revealed that BSPs can show false signals of population decline under several biologically plausible combinations of population structure and sampling strategy, suggesting that the interpretation of several previous studies may need to be re-evaluated. We found that sampling strategies present a trade-off between minimizing the risk of false positives and maximizing the power to detect recent demographic events. Therefore, a balanced sampling strategy whereby samples are distributed over several populations provides the best scheme for inferring demographic change over the time scale that is typically of interest. Case studies demonstrate that although the structure effect should always be accounted for, some systems will be more susceptible than others. Our study provides a much-needed quantification of the structure effect in BSP analyses and some practical guidelines to keep in mind before attempting such analyses.

D23SY30RT16:09R8

DIVERGENCE AND GENE FLOW IN LISSOTRITON NEWTS

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Historical patterns and consequences of genetic exchange between hybridizing species are poorly understood, but new genomic and analytical tools promise a rapid progress. We will present the results of research on historical demography and genome-wide patterns of gene flow between hybridizing newts Lissotriton montandoni (Lm) and L. vulgaris (Lv). These newts are characterized by a long history of genetic exchange, as evidenced by the mitochondrial DNA (mtDNA) data. Multiple, spatially and temporally distinct introgression events from Lv resulted in complete mtDNA replacement in Lm. We used various classes of molecular nuclear markers to obtain information about the extent and historical patterns of gene flow between the two species. Fifteen microsatellite loci and 139 coding SNPs surveyed in multiple populations demonstrated a very limited recent admixture in the nuclear genome. Genome-wide patterns of long-term gene flow were assessed by analyzing of over 300 000 SNPs derived from high-throughput transcriptome sequencing. The inferences based on the joint twopopulation allele frequency spectrum revealed an extensive, asymmetric gene flow from *Lv* to *Lm* and models allowing for demographic changes and temporal variation in the strength of gene flow provided best fit to the data. The species distribution modeling based on contemporary and historical climatic data suggests that temporal variation in gene flow between *Lm* and *Lv* may be linked to range changes the newts experienced during the Pleistocene climatic oscillations. Sequences of over 50 nuclear markers collected throughout the species ranges and analyzed within the coalescent framework provided evidence for genomic heterogeneity in gene flow. Our study demonstrates the complexity of long-term genetic exchange between hybridizing species and the power of new sequencing technologies for a comprehensive characterization of interspecific gene flow in non-model organisms possessing huge genomes.

D23SY30RT16:33R8

MULTI-LOCI BAYESIAN INFERENCE OF THE ANGUILLA SP. GENUS RADIATION AND COMPARATIVE POPULATION GENETICS OF THE MAIN LINEAGES

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Anquilla sp. dispersal and putative spawning areas have divided the scientific community for decades. The rise of the Atlantic species is an unsolved question for which the most supported hypotheses are incompatible: either A. anguilla and A. rostrata came through the Tethys Corridor (closed ~30Mya) or through the Panama Isthmus (open until ~3 Mya). Our study focuses on reconstructing the evolutionary history of freshwater eels. The goal is to ascertain the global demographic dynamics through time for species from the major oceanic basins. Our analyses include TMRCA and diversity calculations, neutrality tests and molecular clock calibration. Using Sanger technology, we sequenced 5 mitochondrial markers, for a total of 304 fish from 10 species. With Bayesian coalescent inference, we estimated genealogies, branch lengths, substitution model parameters and population parameters through time. To infer the timing of demographic events and attach a real-time scale, we used prior information available in the literature. The estimated mutation rate (1.82E-9/bp.yr) and nodes ages were similar to widely accepted reports based on much smaller data sets. However, our results clearly favor the Panama Isthmus route hypothesis over the most popular Tethys route scenario for the settling of the Atlantic eels. Although most species are outside safe ecological limits, we detect genetic signatures of expansion in nearly all sampled populations (10 to 100 fold increases of Ne). These expansions (~400 000 to 720 000 ya) succeeded the mid-Pleistocene transition, a major change in glacial-interglacial cycles. Post-MPT warmth yielded a re-organization of the ocean circulation at intermediate depth and the egress of low-frequency variability in Pacific surface temperatures, impelling a demographic rise. The reported severe decline calls for a better hold of the impact of climate and anthropogenic factors on the resilience potential of these fascinating catadromous fish.

D23SY30RT16:57R8

PHYLOGENY AND EVOLUTION OF THE PLEUROCARPOUS MOSS GENUS ANDOA (HYPNACEAE)

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Andoa is a genus of Hypnaceae, one of the most complex pleurocarpous moss families that is resolved as highly polyphyletic in molecular phylogenetic reconstructions. The extant distribution of the single *Andoa* species, *A. berthelotiana*, in the archipelagos of Azores, Madeira and Canaries classifies it as a Macaronesian endemic. However, fossil records from Continental Europe revealed a larger distribution area in the past. Probably *Andoa* disappeared from Continental Europe during the late Tertiary glaciations and sought for refuge in Macaronesia, where it survived until today. Biogeographic affinities of Hypnaceae in Macaronesia as well as the phylogenetic position and relationships of *Andoa* are still little known. Recent phylogenetic analyses based on chloroplast, nuclear and mitochondrial DNA regions as well as AFLP fingerprinting revealed two geographically separated subgroups within *Andoa berthelotiana*, one comprising plants from Madeira and Canary Islands and the other plants from the Azores. The results also suggest a colonization pattern from the Azores to Madeira and the Canary Islands. Moreover, the closest relatives of *Andoa*, as indicated by the molecular data, were considered to belong to different pleurocarpous families. Other questions remain to be addressed, such as whether the two subgroups relate to 'cryptic speciation' or are also morphologically differentiated.

D23SY30RT17:45R8

PUTTING SOCIAL MICROBES ON THE MAP: A PHYLOGENOMICS APPROACH

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Careful analysis of natural genetic variation is crucial for understanding the processes driving the evolution of phenotypic traits in natural populations of microorganisms, including social traits. These processes include both those that generate new genetic variation (mutation, recombination, horizontal gene transfer) and those that determine the dynamics and ultimate fate of such variation (selection and drift). However, our understanding of natural microbial genetic diversity and how it relates to phenotypic divergence is extremely limited, including for social traits such as those exhibited by the myxobacteria. Based on multi-locus sequence types (MLST), several recent studies have started to characterize the population structure and spatial distribution of the genetic variance in natural isolates of a prominent member of this class of social delta-proteobacteria, Myxococcus xanthus. Here, we extend this work much further by applying a phylogenomics approach that aims at putting microbial genomes on a map spanning metric scales as disparate as 10^{\lambda}-9 m (fruiting bodies) up to 10^{\lambda}7 m (continents). Given the geographic information, we strive to disentangle and quantify the strength of the various evolutionary forces and how these shape the biogeography in this social model organism in the wild.

D23SY30RT18:09R8

PHYLOGEOGRAPHY AND CONSERVATION OF A PROTECTED MOTH

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Phytophagous insects, one of the most diverse animal groups, and their host plants provide excellent model systems to assess the relative contribution of extrinsic environmental factors and intrinsic lineage-specific traits to diversification. We selected the Spanish Moon Moth Graellsia isabellae, an insect that feeds on three pine species, as a model organism to study how modifications in habitat availability influenced the survival of organisms. DNA variation in one mitochondrial and nine nuclear microsatellite markers revealed six clusters across the 28 studied populations in Spain and France. This strong phylogeographical pattern was most likely driven by oscillations in the host-plant distribution range, as four of the six clusters coincide with glacial refugia for Pinus sylvestris, the main host. Both the Central Iberian and French Alpine clusters show the lowest genetic diversity and they are probably the result of postglacial range expansions. We propose the Late Quaternary retraction of P. sylvestris in Southern Spain as the ecological driver for a host-shift to other congeneric species: P. nigra and P. halepensis. Lastly, we define six evolutionary significant units (four of them congruent with the subspecific taxonomy) intimately related to suitable habitat, the main factor shaping the identity of this phytophagous lepidopteran.

D23SY30RT18:33R8

HOW DO DIFFERENT GEOGRAPHICAL BARRIERS AFFECT RING SPECIES FORMATION AND PERSISTENCE?

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Ring species are spatially extended populations that have expanded around a geographical barrier in such a way that the two terminal forms, which meet after many generations, have become reproductively isolated, despite on-going gene flow around the ring. They represent a unique opportunity to study the role of geographical features in speciation. Here we simulate the evolution of ring species using a neutral agent-based model with sexual reproduction, mutation and dispersal. Mating is constrained by a mating area and a critical genetic distance, above which other individuals are no longer considered potential mates. We show that ring species are rare and often unstable to speciation or mixing. However, they can persist for extended times depending on the fine-tuning population, individual, and landscape parameters. The genetic restriction in mating leads to selection against rare types in the area of secondary contact, which contributes to ring species persistence. For the greenish warblers, one of the best documented ring species known to date, our analysis implies that the expanded area near the point of secondary contact is important for extending the duration of the ring. Decreasing the habitable area of the region of secondary contact increases genetic fluctuations there and facilitates the reversal of incomplete speciation, demonstrating the decisive role played by specific landscape features. Our results also demonstrate that geographical barriers can cause speciation even when they do not cause complete isolation.

D23SY30RT18:57R8

A RANGE-WIDE SYNTHESIS AND TIMELINE FOR PHYLOGEOGRAPHIC EVENTS IN THE RED FOX (VULPES VULPES)

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Many boreo-temperate mammals have a Pleistocene fossil record throughout Eurasia and North America, but only few have a contemporary distribution that spans this large area. Examples of Holarctic-distributed carnivores are the brown bear, grey wolf and red fox, all three ecological generalists with large dispersal capacity and a high adaptive flexibility. While the two former have been examined extensively across their ranges, no phylogeographic study of the red fox has been conducted across its entire Holarctic range. Moreover, no study included samples from central Asia, leaving a large sampling gap in the middle of the Eurasian landmass. We obtained the first mitochondrial DNA sequence data of red foxes from Siberia, and new sequences from several European populations. In a range-wide synthesis of more than 500 previously published and publicly available red fox mitochondrial control region sequences and 52 newly obtained sequences, we describe the pattern and timing of major phylogeographic events in red foxes, using a Bayesian coalescence approach with multiple fossil tip and root calibration points. All newly sequenced individuals belonged to the previously described Holarctic lineage. Our analyses confirmed the presence of three Nearctic- and two Japan-restricted lineages that were formed since the Mid/Late Pleistocene. The phylogeographic history of red foxes is highly similar to that previously described for grey wolves and brown bears, indicating that climatic fluctuations and habitat changes since the Pleistocene had similar effects on these highly mobile generalist species. All three species originally diversified in Eurasia and later colonized North America and Japan several times independently. North American lineages persisted through the last glacial maximum south of the ice sheets, meeting more recent colonizers from Beringia during postglacial expansion into the northern Nearctic.

POSTERS

D21SY30PS0011

THE PHYLOGENY AND BIOGEOGRAPHY OF BLENNIIDAE (PERCIFORMES)

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The blenniidae are a diverse family of tropical and subtropical benthic marine fish, with more than 400 species distributed worldwide. We present a novel phylogeny of Blenniidae, based on nuclear and mitochondrial markers, and discuss its taxonomic and biogeographic implications.

D21SY30PS0053

TEPHRITOIDEA (INSECTS: DIPTERA) PHYLOGENY, BASED ON MORPHOLOGICAL AND MOLECULAR DATA

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Tephritoidea family includes families Lonchaeidae, Tephritidae, Ulidiidae (uncluding Otitidae), Platystomatidae, Pyrgotidae, Tachiniscidae, Pallopteridae, Piophilidae and Richardiidae. We analyzed external morphology and genital structures of all these families. We found apomorphies and plesiomorphies in morphological characters and reconstructed modern phylogeny of the group. We discovered several new aspects of the evolution of this group, including that Ulidiidae + Otitidae form a paraphyletic group, referring to derived Tephritoidea. Therefore, we consider Ulidiidae and Otitidae as separate families. We also studied sequences of the COI gene of 58 specimens of 25 species of Ulidiidae and Otitidae. COI was shown to be an effective marker to distinguish Ulidiidae and Otitidae species. Monophyly of the family Ulidiidae with the inclusion of Homalocephala not confirmed on any of the molecular reconstruction. The position of the genus Homalocephala is indefinite. Our study of genital structures has shown that the genus Homalocephala cannot be included neither in Ulidiidae, nor in Otitidae. It represents the separate clade within the Tephritoidea, which requires further study.

PHYLOGENETIC AND MORPHOMETRIC ANALYSIS OF THE SPECIES OF ORESTIAS (TELEOSTEI; CYPRINODONTIDAE) OF THE SOUTHERN CHILEAN ALTIPLANO SUPPORT THE HYPOTHESIS OF A RECENT ORIGIN OF THE SPECIES OF THE LAUCA NATIONAL PARK MODULATED BY THE COLLAPSE OF THE PARINACOTA VOLCANO

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The genus *Orestias* is composed of 44 species distributed along the Altiplano of South America. Seven species have been described for Chile, four of which inhabit Lauca National Park (LNP). We sampled all species in Chile, and also new populations of Orestias within LNP. We constructed a molecular phylogeny using three mitochondrial markers (ND2, Cyt b and the control region). Additionally we performed a geometrical morphometric analysis of body form. The phylogenetic reconstruction showed one lineage (species) for each locality. The exception to this pattern was found in the species described in the LNP, for which a polytomy was recovered. Additionally, a new lineage was found within LNP, composed of localities not previously analyzed. In the morphometric analysis, individuals from Salt pan habitat were differentiated from those from rivers, lakes and wetlands. A second analysis focusing on localities showed that O. ascotanensis and O. aloriae (both of Salt pan habitat) are different among them, and also different from all individuals of LNP. These analyses showed that in the LNP there is little differentiation among the individuals from different localities, and that differences are no attributable to the different environments they inhabit. Based on these results, we infer that the differentiation process of Orestias in the Chilean Altiplano has been mainly allopatric. Additionally, the high morphological and phylogenetic similarity found in the species described in LNP suggests a recent differentiation. This is concordant with geological information, which indicates that the formation in the LNP occurred about 12 kya due to the collapse of the Parinacota volcano. Acknowledgements: FONDECYT 1110243, ECOS-CONICYT C10802, P05-002 ICM.

REVEALING PHYLOGENETIC RELATIONSHIP AMONG SECALE SPECIES

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Genus *Secale* belongs to the grass tribe Triticeae of Poacea family and includes annual and perennial species. In addition to cultivated rye, weedy and wild species also belong to the genus. The genus is quite heterogeneous with annual, perennial (long or short lived), wild, weedy, and cultivated forms. Taxonomy of the genus is still controversial due to the lack of agreement about diagnostic criteria and the number of species classified in the genus varies from three to 14 in different studies. According to most widely accepted classification, the taxon contains four species: *Secale cereale*, *Secale vavilovii*, *Secale sylvestre*, *Secale striticum*. Among these, *Secale cereale* and *Secale striticum* are polytypic. Furthermore there is no consensus on the origin of cultivated rye and evolutionary history of genus *Secale*. In order to gain new insights about phylogenetic relationships among *Secale* species and the degree of genetic diversity of genus, 142 different accessions of *Secale* species from different ecogeographical origins, with a detailed focus on Turkey and the Fertile Crescent is going to be evaluated morphologically and molecularly. Molecular analyses will include SSR and mitochondrial, chloroplastic and nuclear SNP.

COMPARATIVE TRANSCRIPTOMICS OF EARLY DIPTERAN DEVELOPMENT

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We are carrying out a comparative analysis of transcriptomic sequence data in early embryos of three emerging experimental dipteran model systems. *Clogmia albipunctata* belongs to a lineage of flies believed to have diverged early in the evolution of the dipterans, while the lineage leading to *Megaselia abdita* branched intermediately, at the base of the cyclorraphans, and the lineage leading to *Episyrphus balteatus* diverged later in the cyclorraphans. We have acquired and assembled transcriptomic sequences at early embryonic stages in *Clogmia albipunctata* and *Megaselia abdita*. We compare these sequences to those from *Episyrphus balteatus* as well as transcriptomic and genomic sequences from drosophilids and/or mosquitoes. These datasets form the basis of a new phylogenomic assessment of dipteran relationships. It places psychodid moth midges (such as *Clogmia*) as a sister group to the brachycerans, rather than within the culicomorph lineage, in contrast to another recent study (Wiegman et al., 2011). Furthermore, we have analysed patterns of gene duplication in our datasets. Finally, we have verified information present in our transcriptomes by manual curation, in situ hybridization, and verification of alternative splicing events among a subset of genes present in the data.

SYMPATRIC SPECIATION IN THE GULF OF GUINEA ISLANDS FINCHES IS SUPPORTED BY MULTIPLE INDEPENDENT MOLECULAR MARKERS

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Sympatric speciation is contentious and convincing examples from nature remain very scarce. Here we present multiple lines of molecular evidence supporting a sympatric origin for one of the most enigmatic birds in the world, the São Tomé grosbeak *Crithagra concolor*. This endemic from São Tomé Island, Gulf of Guinea, was initially placed in a monotypic genus, *Neospiza*. Mitochondrial data placed it as sister to the Príncipe seedeater *Crithagra* rufobrunneus, a Gulf of Guinea endemic with populations on the islands of São Tomé, Príncipe and Boné de Jóquei. For 29-30 seedeaters (9-10 per population), three grosbeak samples and outgroups we sequenced mitochondrial markers (c. 1000 bp), 27 nuclear introns and 6 exons (c. 20,000 bp), and we genotyped 33 microsatellite loci. All molecular markers inferred a closer relationship between the São Tomé grosbeak and the seedeater population from São Tomé, than between all three allopatric seedeater populations. Mitochondrial markers had a strong phylogenetic signal (no shared haplotypes, c. 2.5% sequence divergence between the sympatric grosbeak and seedeater). In contrast, lineage sorting of the nuclear markers was incomplete (albeit unique grosbeak mutations were also present). This pattern is consistent with sympatric speciation followed by incomplete lineage sorting of the nuclear markers rather than with past hybridization events (that would have homogenised the mtDNA faster than the nDNA).

MORPHOLOGICAL CONVERGENCE AND DISPARITY IN MALAGASY TENRECS (AFROSORICIDA, TENRECIDAE)

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Understanding patterns of variation in morphological diversity remains a central challenge in evolutionary biology. Two aspects of morphological variation which attract particular interest are convergence - the independent evolution of similar morphologies in phylogenetically distant speciesand disparity –the range or significance of morphology in a given sample of organisms. Tenrecs appear to be both disparate from each other and convergent with other species such as hedgehogs, shrews, moles and otters. It has been suggested that these morphological patterns may arise as a function of ecological similarities and differences among the species. However, previous studies have neither quantified the degree of convergence or disparity present in tenrecs nor attempted to identify reasons for the occurrence of these patterns. These omissions represent significant gaps in our understanding of the evolution of morphological diversity. Theoretical models indicate that some degree of convergence is expected in most phylogenies. Quantitative analysis is therefore crucial if we are to identify significantly unusual and interesting patterns of convergence.
We collected morphological data from skulls and limbs of both tenrecs and the mammals they convergently resemble. Using a combination of both traditional and geometric morphometric techniques, we assess the evidence for significant morphological disparity among tenrecs and significant convergences among tenrecs and other mammal species. We also compare species' ecological niches using novel techniques and determine the significance of ecological similarity in contributing to observed patterns of morphological convergence. The methods developed in this work – quantification of previously subjective morphological patterns and assessment of correlations between ecological and phenotypic similarities – have useful applications in diverse research areas including functional diversity biodiversity monitoring and conservation.

CONNECTIVITY IN FRAGMENTED MARINE ENVIRONMENTS: A COMPARATIVE APPROACH BETWEEN METHODS IN TWO LOW DISPERSIVE SPECIES

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Marine populations are considered as demographically open populations linked by strong connectivity. The low levels of genetic differentiation observed at large geographic distances in many species is explained by the high dispersal capacities of these organisms linked with the occurrence of a larval phase in their life cycle. However, this view was recently questioned in various species such as sessile cnidarians in which significant genetic structure has been reported at local and regional scales. These results call for a reassessment of the connectivity in the sea by focusing on low dispersive species. In order to disentangle the relative impact of demographic and environmental processes on their connectivity, we performed a comparative analysis between two octocorals, the red coral Corallium rubrum and the red gorgonian Paramuricea clavata, in the same geographical area (North-Western Mediterranean). These species both inhabit contrasted ecological environment (e.g. different thermal regimes) and are confronted to past and current climate changes. Based on an extensive sampling and on microsatellite markers, we first compare different methods allowing the analysis of genetic structure and dispersal at different time scales: individual clustering, assignment, isolation by distance and probabilistic estimates of long-term dispersal. Then, we interpret genetic estimates of dispersal in the context of the evolution of these species in their complex landscape and of the interaction between migration and selection. The obtained results enhance our understanding of the mechanisms implied in the short distance differentiation observed in these species, including migration rate, effective population size and environmental parameters. We finally discuss the management and conservation issues related to these results and propose new research perspectives in this context.

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ANALYSIS OF POPULATION STRUCTURE OF GUPPIES IN NORTH-EASTERN VENEZUELA, THE AREA OF PUTATIVE INCIPIENT SPECIATION

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Geographic barriers to gene flow are one of the main causes of genetic isolation. Reproductive isolation often follows, which in turn may lead to speciation. The guppy from the Cumaná region of Eastern Venezuela has been previously described as a case of incipient speciation, significantly differentiated from the common guppy, *Poecilia reticulata*, which occurs on the other side of Cordillera de la Costa in streams belonging to the Orinoco drainage. Here, we present a comprehensive analysis of differentiation between guppy populations in these two regions of Eastern Venezuela based on mtDNA and a set of 15 microsatellites. We also analyse four phenotypic traits which have been found in earlier work to differ between the Cumaná and common guppy. Guppies inhabiting the two regions divided by Cordillera de la Costa indeed show significant genetic differentiation, but most genetic variation was distributed among populations within regions. Most guppies in the Cumaná region had mtDNA from a distinct lineage, but we also found evidence for a widespread introgression of mtDNA from the Orinoco drainage into the Cumaná region. Populations from the two regions differed significantly in the number of black crescents, but not in other phenotypic traits analysed. Overall, our findings are consistent with only partial genetic isolation between the two geographic regions. Gene flow from the Orinoco drainage may have caused the loss of typical Cumaná morphology in many populations.

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SPECIATION BY DEPTH IN AN OCEANIC FISH?

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The glacial and post-glacial processes of the Pleistocene have been recognised to play a major role in promoting genetic differentiation, and in shaping the present-day diversity and distribution of biota. Little is known about the impact that such events had on processes of divergence in geographically overlapping, depth-stratified populations. In the ocean, diversification along depth gradients is likely to alter the visual environment of organisms, and adaptive changes in visual systems may be expected. *Sebastes mentella*, a now highly exploited deep-sea fish, exhibits patterns of substructure along a depth gradient in the central North Atlantic; here we performed a molecular genetic investigation of samples from fish collected above and below 500m, using the mitochondrial control region and the gene coding for the visual-pigment rhodopsin. We provide evidence for potential adaptation to different depths, by detecting a specific amino acid replacement, within the transmembrane domain of the rhodopsin gene. Mitochondrial DNA results reflect a scenario of long-term demographic independence between the *S. mentella* groups and raise the possibility that these 'stocks' may in fact be two incipient species. We stress the importance of incorporating depth as a diversifying agent, when assessing biodiversity and population structure in oceanic species.

EFFECTS OF CONNECTIVITY AND SIZE OF LOCAL SITES ON SPECIES DIVERSITY-GENE DIVERSITY CORRELATIONS WITHIN (META)COMMUNITIES : INSIGHTS FROM NEUTRAL THEORIES

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In the last decade, there has been a growing interest in the simultaneous study of genetic diversity within populations and species diversity within associated communities. It has been argued that "neutral processes" such as drift and migration should similarly affect both organizational levels, thus contributing to positive covariation of diversities among sites, the so-called species-genetic diversity correlations (SGDCs). However, environmentally dependent competition and ecological characteristics of species may weaken, or even reverse, such SGDCs. Empirical studies have revealed both negative and positive SGDCs over a wide array of ecological situations, leading to discussions over the relative strength of "neutral" against "niche" processes in population / community construction. However, a proper quantitative neutral model including both levels has yet to be produced as a null model for SGDCs. We propose a unifying mainland-islands model with a sampling theory to explore the effects of sites' connectivity and size, as well as the importance of intra-specific neutral mutation rate, on SGDCs. We show that variation in size and connectivity among sampled sites contributes to positive SGDCs through the relative strength of drift compared to migration. Moreover neutral gene mutation can significantly affect SGDCs, leading to negative correlations under realistic situations. We therefore argue that neutrality should not necessarily generate positive SGDC and provide some general guidelines for interpreting such joint patterns of diversity.

ECOLOGY AND SEXUAL SELECTION: THE EVOLUTION OF WING PIGMENTATION AND SEXUAL DIMORPHISM IN CALOPTERYGID DAMSELFLIES

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Sexual selection is thought to lead to sexual dimorphism, following a transitional stage where both sexes have similar phenotypes due to a shared genetic architecture. In addition, sexual selection might facilitate both speciation and possibly also extinction. However, our knowledge about how the environment shapes sexual selection regimes and how ecology and sexual selection interact is still limited. Here we report the results from a study on the evolution of wing pigmentation in calopterygid damselflies. We investigated the effects of sexual selection on sexual dimorphism, speciation and extinction and the possible thermoregulatory consequences of wing pigmentation using a mixture of phylogenetic comparative analyses, field data and experiments. First, we traced the evolution of wing pigmentation and reconstructed ancestral states of male and female phenotypes. Our results indicate that clear wings are the ancestral state, that pigmentation is costly to females and sexual selection results in sexual dimorphism. We further demonstrate that pigmentation elevates speciation rates and tends to also elevate extinction rates. We document a significant biogeographic association with pigmented species primarily occupying northern temperate regions with cooler climates. Field observations and experiments on two temperate sympatric species suggest an interaction between pigmentation, thermoregulation and sexual selection, although body temperature is also likely to be influenced by other phenotypic traits such as body mass, microhabitat selection and thermoregulatory behaviors. Taken together, our results suggest an important role for wing pigmentation in speciation and sexual selection in males, but with a net cost to females. However, wing pigmentation does not necessarily increase ecological adaptation and species longevity, and its primary function therefore lies in sexual signalling and species recognition.

GLACIAL AND POSTGLACIAL HISTORY OF BETULA HUMILIS IN POLAND AND BELARUS

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Trees and shrubs of the Betula genus inhabit diverse habitats in the Northern Hemisphere. In Europe, there are two tree birch species: Betula pendula and B. pubescens, and two shrub birches: B. nana and B. humilis. Both palaeontological and genetic investigations have shown that B. pendula and B. pubescens could have survived the Last Glacial Maximum (LGM) not only in southern European refugia but also at higher latitudes and after Holocene warming a rapid recolonisation occurred. The boreal shrub *B*. *nana* could have also existed in a periglacial area during the last glaciation. It is very difficult to reconstruct the glacial and postglacial history of *B. humilis* using solely palaeoecological data because the macrofossil record is rather scarce, and the pollen of this species is classified as B. nana type. The aim of the present study is to analyse variation of chloroplast DNA (cpDNA) haplotypes of *B. humilis* in order to describe the glacial and post-glacial history of the species in Poland and Belarus. Investigations were conducted in 25 populations of *B. humilis*. In total, 21 haplotypes were found but only two of them occurred with high frequencies. Clear predominance of two haplotypes in the studied area could be explained in terms of recolonisation from two distinct refugia. A weak genetic structure within the area populated by *B. humilis* has suggested that the species could have survived the LGM at higher latitudes. A glacial refugium of *B. humilis* was probably located in eastern Europe, another one could be situated in the Alps or the Carpathians. Low level of genetic diversity has not confirmed a hypothesis about *B. humilis* glacial isolate in south-eastern Poland. In turn, high haplotypic diversity has been revealed in some populations of *B. humilis* in north-eastern Poland and central Belarus. This pattern can likely be explained as a contact zone between different phylogeographic lineages.

PHYLOGEOGRAPHIC STUDIES REVEAL DIVERSE SPECIATION PATTERNS IN CENTRAL AFRICAN TROPICAL HERBS (MARANTACEAE)

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Vegetation history in tropical Africa is still to date largely unknown. Hypotheses include major speciation events during Pleistocene climate changes and ecological speciation across abiotic gradients. Phylogeographic studies might shed new light on this matter as indicated by similar studies in temperate regions; the challenges in tropical Africa being different highly diverse biotic and abiotic conditions and a distinct climatic and geological history. Here we analysed phylogeographic patterns (chloroplast-DNA sequences, AFLP, microsatellites) of eight widespread herb and liana species from four different genera of the family Marantaceae (Zingiberales) including four pairs of sister species. Across all investigated species, intraspecific patterns of genetic diversity from chloroplast DNA were concordant with overall Marantaceae diversity: higher in topographically rich Gabon and showing traces of expansion across the rather flat central Cameroon and the Congo basin. However, comparing sister species relationships between different genera revealed different levels and modes of speciation. Patterns in the genus Haumania suggest allopatric speciation with genetic pattern concordant with postulated Pleistocene refugia followed by current intraspecific gene flow in overlapping distribution areas. Concordant pattern of genetic diversity and the absence of hybridization in the genus Megaphrynium might hint towards sympatric speciation through adaptation to different pollinator species. In the genus Marantochloa allopatric speciation followed by complete incompatibility between sister species is indicated in one species pair. In a second case two morphologically distinct sister species are indistinguishable genetically and the mode of speciation still open to debate. This study gives an insight into the diverse and ongoing evolutionary processes in central tropical Africa and the role of different geographic areas and ecological traits in the evolutionary process.

30. Phylogenetics and Phylogeography

D21SY30PS0432

A PHYLOGENY OF SUBTRIBE FERULINAE (TRIBE SCANDICEAE, APIACEAE) INFERRED FROM NUCLEAR RDNA ITS AND CPDNA (*RPS*16, *RPOC*1 INTRONS AND *RPOB-TRNC* SPACER) SEQUENCE VARIATION

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Phylogenetic relationships within Apiaceae tribe Scandiceae subtribe Ferulinae were investigated using sequence data from the chloroplast DNA and nuclear ribosomal DNA internal transcribed spacer (ITS) regions. One hundred and seventy-six accessions were examined, representing the genera of the subtribe (Ferula, Dorema, Leutea) and outgroup taxa. The cpDNA markers included three non-coding loci: *rpoB-trnC* intergenic spacer, *rps*16 intron and *rpo*C1 intron. These loci were analyzed separately and combined to assess their relative utility for resolving relationships. Partition homogeneity tests yielded trees that demonstrated the incongruency between cpDNA and nrDNA regions. Among the loci examined, *rps*16 intron showed less variability relative to its size than the other used cpDNA markers. Phylogenies derived from maximum parsimony and Bayesian analyses of combined cp and nr DNA sequences for 156 accessions (representing 114 species of Ferula, 6 species of Dorema, 9 species of Leutea and 10 outgroup taxa) were resolved to some extent. Based on these analyses, Dorema is nested within *Ferula* and should be therefore subsumed within the latter. In contrast, *Leutea* appears to be in a sister position to *Ferula* and should be retained as a separate genus. Subtribe Ferulinae is diversified in central Asia, floristically encompassing the central and eastern part of the Irano-Turanian Floristic Region where the representatives of nearly all major groups occur, with the notable exception of two groups that are mostly distributed in the Mediterranean region and in China. However, since the species boundaries in *Ferula* are not clear, one may also suspect that the high number of species, particularly of the central Asiatic endemics, may have resulted from taxonomic splitting rather than rapid radiation of the genus in this region.

GENETIC STRUCTURE OF AN ALPINE SHRUB RHODODENDRON FERRUGINEUM (ERICACEAE)

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Genetic variation within plant species is determined by a number of different factors such as reproductive mode, breeding system or life history traits. In alpine regions, the combined environmental changes across altitude and longitude can result in complex local patterns of genetic variation. Due to the uncertainty of plant responses to climate change, an understanding of current distribution patterns of population genetic diversity is fundamental for establishing management strategies for resilient species. Pyrenees are a mountain range stretching across the isthmus that lies between the Iberian Peninsula and the rest of the European continent. Acting as a biogeographical barrier, they are the southern limit of numerous species. Both influences of the Mediterranean Sea (east) and the Atlantic Ocean (west) allowed refuge zones during the glacial periods. This can lead to specific genetic pattern of Pyrenean species. This work focuses on R. ferrugineum which is an alpine shrub distributed in the Pyrenees, Alps and Jura. R. ferrugineum is an entomogamous species with a mixed mating system. To assess the genetic structure within and among populations we genotyped individuals from 33 populations (647 ind.) at 27 microsatellites loci. We found a structuration with 5 genetic clusters along a longitudinal gradient across the Pyrenees. This structuration is supported by Mantel tests that revealed a positive relationship between genetic distance matrix and geographic distance matrix across populations located on the Pyrenees. Populations from the Alps and Jura formed an additional cluster genetically close to the easternmost Pyrenean populations. Therefore, Pyrenean populations may derive from Alpine populations and appeared to have colonized the Pyrenees from east to west (gene flow analyzes). The high structuration across the Pyrenees may be explained by a strong level of clonality and autogamy in this species but also by some historical and colonization events.

DEEP PHYLOGENETIC SIGNAL IN THE DISTRIBUTION OF ANGIOSPERM DIVERSITY ALONG ELEVATIONAL GRADIENTS IN A MEDITERRANEAN BIODIVERSITY HOTSPOT

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Quantifying change in species and phylogenetic composition across biogeographic regions or environmental gradients (β-diversity and β-phylodiversity respectively) provides insight into the ecological and evolutionary processes that shape biodiversity. Here we investigated the entire angiosperm flora of the mountains of the Baetic range in Andalusia, one of the most diverse regions within the Mediterranean Basin Biodiversity Hotspot, quantifying β-phylodiversity between bioclimatic elevational belts while accounting for differences in species β-diversity. We also measured phylogenetic structure within each bioclimatic elevational belt, to assess how phylogenetic composition changes along the elevational gradients. We found β -phylodiversity turnover was very strong among elevational ranges both within and between Sierras, but was significantly lower between floras of the same elevational range from different Sierras. This pattern was strongest within the Core Eudicots, but broke down in the Monocots and ancient Dicots. At medium to high elevations the flora was phylogenetically clustered, but was random with respect to phylogeny at low elevations. Once again this differed between the Core Eudicots and Monocots-ancient Dicots when analysed independently, the former being random with respect to phylogeny at all elevational ranges and the latter being clustered at high elevations. These results indicate that deep phylogenetic signal is driving the spatial distribution of angiosperm plants along elevational gradients in the Baetic mountains, and suggests that species occuring at high elevations, and thus likely more vulnerable to climate change, are non-random with respect to phylogeny.

USING NEXT-GEN SEQUENCING TO RESOLVE THE PHYLOGENY OF TROPICAL TREE FAMILY

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Next-Generation Sequencing have brought a great leap forward in a large range of biological questions, including phylogenetics. However, the usual genome reduction or organellar purification steps are often time-consuming, costly, and require large amounts of biological material. This is especially problematic in tropical woody plants where secondary compounds are likely to interfere with chemical reactions related to DNA extractions and library preparation. We explored whether shallow shotgun sequencing of total DNA may be used to resolve the phylogeny of the pantropical tree family Chrysobalanaceae. This technique takes advantage of the difference in the per-cell-copy number between organellar, nuclear ribosomal and nuclear DNA, and thus does not require any treatment of the extracted total DNA prior to the library preparation. Even with a very low sequencing depth of the nuclear DNA, we were able to assemble the total plastidic genome (about 160 kb) and the nuclear ribosomal region (around 6.5 kb), and we retrieved almost all the mitochondrial genes (around 70 kb) for all the Chrysobalanaceae species tested (n = 8). Using these data, we resolved ancient nodes of the family phylogeny that had remained unresolved in a previous Sanger-based study. We also successfully demonstrate that this technique may be applied to two other tropical tree families also known for being rich in secondary organic compounds. In this study, we demonstrate that shallow sequencing of total DNA is efficient to build reliable phylogenetic hypotheses, especially in chemically complex organisms such as tropical plants. This approach should help unveil key remaining on the origin and diversification of flowering plants.

30. Phylogenetics and Phylogeography

D21SY30PS0503

AN ISLAND CALLED INDIA: PHYLOGENETIC PATTERNS ACROSS MULTIPLE TAXONOMIC GROUPS REVEAL ENDEMIC RADIATIONS

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The Indian subcontinent is an interesting biogeographical entity as it is isolated from the rest of the Asian landmass by very high mountain ranges like the Himalayas in the north and is surrounded by ocean in the south. Furthermore much of this landmass was part of a Gondwanan fragment that merged with Asia around 35-55 mya. Thus Indian subcontinent has witnessed prolonged periods of isolation and continues to be largely cut off from much of Asia. In this regard the subcontinent is very similar to an island separated from the mainland (Asia). One interesting feature of an isolated island is the presence of endemic radiations, i.e., unique clades of taxa whose members are endemic to the island. Recent phylogenetic studies have revealed that India also harbors numerous endemic radiations and these endemic assemblages were generated through in-situ diversification. Thus to this day India is very similar to an isolated island. It is interesting to contrast the Indian scenario with Madagascar an island with which India shares a geological past. Furthermore these finding have important implications for ecological and evolutionary studies on Indian biota.

30. Phylogenetics and Phylogeography

D21SY30PS0512

GENETIC DIVERSITY OF A RELIC MOOSE POPULATION, ALCES ALCES IN THE BIEBRZA VALLEY, NE POLAND

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Moose (Alces alces) are highly mobile mammals that occur across arboreal regions of North America, Europe and Asia. Hundertmark et al. (2002) identified three distinct mtDNA lineages of moose in these regions. The distribution of these lineages indicated a geographic structure associated with a glacial history. To examine the genetic diversity and population structure of moose in northeastern Poland we analyzed mtDNA sequences of 1747 bp long (control region combined with cytochrome b) of 500 individuals from 13 populations. Our study revealed presence of 12 haplotype belonging to the European lineage, from which only five haplotypes were early detected by Hundertmark et. al. (2002) exclusively among moose from Scandinavian Peninsula. The haplotype (h) and nucleotide (π) diversity values for analyzed moose samples were respectively 0,76 and 1,2%. Altogether, 27 sites were variable and all substitutions appeared to be transitions. In our study the haplotype H1 was mainly present in the Biebrza Valley and it was divergented from the remaining European haplotypes. Phylogenetic relationships were computed using a Neighbor-Joining (NJ) and a Maximum Likelihood (ML) trees. In both analysis the clade contained the unique haplotype H1 from the Biebrza Valley was the most distinct within the European lineage. Net sequence divergence between these clade and the others European haplotype amount 1,4%, which means moose from the Biebrza Valley separated 23000 years ago, eg during the Last Glacial Maximum (LGM). We also found that genetic differentiation between several moose populations in northeastern Poland was great, for example the Φ ST value between two neighbouring populations: from the Biebrza Valley and the Augustowska Forest was 0.55. In addition, we analyzed others classes of molecular markers: 11 microsatellite loci and markers located on Y chromosome (SRY and YCATs) which also confirmed relic character of moose from the Biebrza Vallev.

I HAVE A GUT FEELING: PUTATIVE SULFIDE-DEPENDENT DISTRIBUTION OF BACTERIAL MOLLICUTE GUT SYMBIONTS IN NIPHARGUS AMPHIPODS

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Chemosynthetic symbioses were first discovered in the 1970s at hydrothermal vents in the deep sea. Subsequently, they were found to occur in a wide variety of marine environments, ranging from shallow-water coastal sediments, wood and whale falls, to cold seeps. Only recently a chemosynthetic symbiosis was identified in a freshwater habitat. An ectosymbiosis between an invertebrate host (Niphargus sp.; Amphipoda) and sulfur-oxidizing bacteria of the genus Thiothrix was identified in an Italian sulfidic cave system. These caves are isolated from surface photosynthetic primary production and demonstrate many geochemical and ecological similarities to marine vents and seeps. Potential symbioses in these freshwater environments in general and with *Niphargus* sp. as the host in particular, have not been fully explored. Here we show the occurrence of gut symbionts of the bacterial class Mollicutes in European freshwater Niphargus amphipods. By sequencing 16S rRNA gene clones of Niphargus gut DNA extract, we identified three clades of Mollicutes, which are closely related to mollicute bacteria found in the gut of the marine hydrothermal vent shrimp *Rimicaris exoculata*. It has been hypothesized that these bacteria might play a role in host nutrition or detoxification, although to date there is no direct evidence. We are exploring the presence/absence pattern of the mollicute bacteria in the gut of various Niphargus species from multiple European cave systems in relation to geochemical parameters, such as sulfide levels. If mollicute gut symbiont presence correlates with a sulfidic host habitat, both in marine and freshwater habitats, a role of the mollicute bacteria in either nutrition or detoxification of their hosts seems likely.

RADIATION PATTERNS WITHIN THE ENDEMIC GENUS PERICALLIS: A MODEL TAXON FOR CONTRASTING PATTERNS OF DIVERSITY BETWEEN MACARONESIAN OCEANIC ARCHIPELAGOS

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Macaronesia is a biogeographical region comprising the volcanic, oceanic archipelagos of the Azores, Madeira, Canary Islands, Salvage Islands and the Cape Verdes, in the Atlantic Ocean. The region is a European Biodiversity hotspot, hosting a rich endemic flora, but there are marked disparities in levels and patterns of endemic diversity across the region. The 'Azores Diversity Enigma' refers to the paucity of single island endemics (SIEs) and in situ radiations in the Azores flora compared to all other oceanic archipelagos. The Macaronesian endemic *Pericallis* is a unique example of a flowering plant genus restricted to the Azores, Madeira and Canary Islands. It has diversified in the Canary Islands with 11 restricted SIEs whereas in the Azores just one species with two ecologically distinct subspecies are recognised. As such, the species diversity patterns closely reflect the disparities evident between the floras of the archipelagos more generally. I have carried out morphological and AFLP analyses, contrasting diversity patterns of *Pericallis* between the Azores and Canary Islands. This talk presents the results of these analyses and discusses how these results can help to explain the 'Azores Diversity Enigma'.

30. Phylogenetics and Phylogeography

D21SY30PS0553

PHYLOGENY OF PALAEARCTIC CALLOPHRYS (LEPIDOPTERA, LYCAENIDAE) BASED ON MOLECULAR DATA, MORPHOLOGY AND BIOLOGY

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Palaearctic Callophrys (Lepidoptera, Lycaenidae) is taxonomically complicated genus. External similarity, some aspects of biology and localization in out-of-the-way regions which are the pecularities of the most species had obstructed investigation of this group. Complex analysis of COI-gene, morphology and some data on biology illuminates problems in systematics and lets to investigate phylogeny and main evolution trends of the group. Morphological analysis shown that the Palaearctic region is inhabited by 15 species of *Callophrys*, 3 of them were described as new by us. The center of biodiversity of the group is situated in Armenian Highland and Iranian Plateau. Morphological analysis let divide the genus into five main groups: C. avis, C. rubi, C. paulae, C. suaveola and C. mystaphia – species groups. Members of these groups have unique combination of external morphological characters and characters of genital structures. This division also correlates with host plant preferendum of each group. Molecular analysis based on COI-gene supports such division but usage of this gene as a barcode marker is impossible due to absence of gap between intra- and interspecific genetic differences in *C. rubi* – group. Probably it's a consequence of relatively late divergention of the group. Phylogenetic scheme lets us suppose that C. avis – group (which includes one species, morphologically close to Nearctic Callophrys) is the basal, and C. suaveola – group is the most advanced. Thus we can define apomorphies and plesiomorphies in morphological characters and in modes of ecological strategies which are as follows: developed lateral lobes of antrum in female genitalia, developed anal lobe of hindwing, developed postdiscal row of spots and usage of Fabaceae as host plants (plesiomorphies); reduced lateral lobes of antrum, rounded hindwing, reduced postdiscal row of spots and usage of Umbelliferae and some Polygonaceae (Rheum spp.) as host plants (apomorphies).

CONSTRUCTING SUPERMATRICES FROM FUNGAL INSD DATA – PROBLEMS AND SOLUTIONS

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Many questions in macroevolution and macroecology are addressed using phylogenies. In many cases large phylogenies, including hundreds to many thousands of taxa, are needed to resolve these questions. The accumulation of sequences from different gene sources in public sequence databases, such as the International Nucleotide Sequence Databases (INSD; GenBank, ENA, DDBJ), has enabled an opportunity to reconstruct large supermatrix phylogenies. Supermatrix and supertree methods have been developed to infer phylogenies from mosaics of data accumulated from disparate studies. However, gene sequence data in INSD are coupled with several challenges regarding sequence quality, missing metadata, incongruity within metadata fields, and taxonomic confusion. One problem is the lack of standardized annotation from which region of the genome the sequence represents. Here we use HMMER to separate sequences from different genes and demonstrate how HMMER can be used to cut out genes/DNA regions within a single sequence. By partitioning sequences it is possible to fit separate substitution models to each gene. It is also possible to remove problematic regions. This is particularly useful in fungi where nuclear ribosomal DNA, characterized by substitution rate heterogeneity, is the most commonly sequenced region. Another problem is dubious taxonomic annotations. Erroneous taxonomic annotations may link sequences from different species together and create a chimeric concatenated sequence. To address this problem we introduce an individual based approach, combined with clustering, that links genes into operational taxonomic units and compare these to a species annotation based approach. The success of this approach depends on the structure of the data and the prevalence of metadata to define individuals. Our methods have been implemented in a software pipeline that can produce either supermatrices or trees for supertree approaches.

PHYLOGENETICS AND SPECIES DELIMITATION IN THE ORCHID GENUS BRASILIORCHIS

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Species are fundamental units of biodiversity. In recently or rapidly divergent clades species limits are often blurred and comprise fascinating opportunities for speciation studies. The absence of clear species boundaries also hampers the development of studies in others areas in biology and is an obstacle to conservation. The orchid genus Brasiliorchis currently contains 14 species mainly from the Brazilian Atlantic Forest Biome. Most morphological diagnostic characters among species are continuous and thus not useful for species identification. Our goals were to contribute to the identification of evolutionary distinct lineages within genus and assess if phylogenetic patterns of diversification agree with a previous morphometric study available. We used sequence data from plastid (psbj-petA, atpl-atpH) and nuclear (ITS1-2) regions considering wide morphological and geographical sampling variation. We sampled 96 individuals, 38 populations and 11 currently recognized species. Parsimony and bayesian analyses were performed considering data separately and concatenated. Two species, B. schunkeana and B. barbozae, were recovered as monophyletic, in agreement with morphological diagnostic characters. The concatenated analysis also suggested B. gracilis to comprise a distinct lineage, corroborating morphometric data. B. kautskyi and B. consanguinea, which are morphologically clearly distinct, are embedded in a poorly resolved clade including the remaining species (B. chrysantha, B. marginata, B. phoenicanthera, B. picta, B. porphyrostele, B. ubatubana), which are not supported by our results. Additional markers and species tree analyses are in progress to clarify diversification patterns in this group.

30. Phylogenetics and Phylogeography

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WEST VS EAST MEDITERRANEAN SEA: ORIGIN AND GENETIC DIFFERENTIATION OF THE SEA CUCUMBER HOLOTHURIA POLII

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We studied the genetic structure of the sea cucumber *Holothuria (Roweothuria) polii* (Delle Chiaje 1823) by analyzing the mitochondrial DNA variation in a fragment of cytochrome oxidase I (COI) and 16S genes. Individuals were collected in 7 locations along the Mediterranean Sea, which cover a wide range of the species distribution. We found high haplotype diversity for COI and moderate for 16S and low nucleotide diversity for both genes. Our results for the COI gene showed many recent and exclusive haplotypes with few mutations between them, which suggests populations might be under expansion. The Western and Eastern Mediterranean populations exhibited slight but significant genetic differentiation showing the highest genetic diversity on the East. The most ancient haplotype was not present in the most western sampling location (Spain). Also, Kusadasi (Turkey) displayed the oldest expansion time, which corresponds to mid Pleistocene epoch, as well as the highest genetic diversity (number of exclusive haplotypes, polymorphisms, haplotype and nucleotide diversity). This suggests that individuals from Kusadasi could be the origin of the subsequent colonizations through the Mediterranean Sea. However, for future research, nuclear markers should be used to confirm this hypothesis. Keywords: Genetic structure, colonization, *Holothuria polii*, Mitochondrial DNA, Mediterranean Sea

WORLDWIDE COLONISATION HISTORY OF THE SEABIRD TICK IXODES URIAE AND WHAT IT CAN TELL US ABOUT THE EVOLUTION OF HOST SPECIALISATION

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Historical patterns of dispersal and population isolation are key components shaping contemporary genetic diversity across landscapes. Here, we investigate the colonisation history of a common ectoparasite of colonial seabirds, the tick *Ixodes uriae*. This tick has a circumpolar distribution across both hemispheres, but has repeatedly formed host-specific races within different seabird host communities. By combining mitochondrial and nuclear data and analyzing this data using both phylogenetic and phylogeographic frameworks, we infer how this species reached its present-day distribution and how the colonisation process has affected the geographic structuring of this tick among colonies and host species. We show the existence of four/five genetic groups that correspond to well-defined geographic regions. Our data indicates that *Ixodes uriae* colonised the southern hemisphere before moving into northern latitudes. However, no relationship between the degree of host race evolution and colonisation history was evident suggesting that host specialisation can evolve rapidly but never leads to speciation. We discuss the possible historical and contemporary mechanisms of large scale dispersal for this ectoparasite and how its biological characteristics may condition current patterns of genetic diversity.

EVOLUTION OF HETEROSTYLY ACROSS THE PHYLOGENY OF SOUTH AFRICAN FLAX SPECIES (*LINUM*, *LINACEAE*)

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Linum (Fam. Linaceae) is a subcosmopolitan genus, which includes about 250 species, and it is widely distributed in temperate and subtropical regions. The genus is organized in five taxonomic sections: Linum, Cathartolinum, Syllinum, Dasylinum and Linopsis. While all the sections are relatively well represented in the North of Africa, Linopsis appears to be the only section represented in the Southern Africa, and it is particularly abundant (in number of species) in South Africa (mostly in the Cape Region). South African Linum is represented by 14 species displaying a comparatively low variation in floral and vegetative traits, which has made difficult taxonomic treatment. Heterostyly has been known in Linum for long, but it was supposed to be restricted to Old World northern hemisphere. Our field sampling has shown that heterostyly and monomorphism is present in some populations and species in South Africa. Linum species of Cape Region are related with South American lineages where heterostyly is absent, rather than to Mediterranean *Linum, where this style polymorphism is common. We explore the extent to which heterostyly is a newly acquired trait in this southern lineage, and what are the ancestral and derived conditions within the variability of traits related to heterostyly shown by this small group of taxa. To do that, we present data on cpDNA and nuclear sequences from 50 populations of this group of species from South Africa and reconstruct trait evolution on the recovered phylogenetic relationships.

NOT BOTHERED BY YOUR SPERM: POSITIVE SELECTION IN A GAMETE RECOGNITION GENE IS NOT DRIVEN BY REINFORCEMENT IN TWO CONGENERIC SEA STARS

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The relative strength of sexual selection and sexual conflict can have important implications for the diversification of gamete recognition proteins and reproductive divergence among populations. However, the selective processes that operate within species are often difficult to discriminate from selection that operates among species, such as reinforcement. Here, we examine patterns of molecular evolution in the male-expressed gamete compatibility gene, bindin, in two broadcast spawning species of sea stars across their partially overlapping geographic range in the eastern Pacific. We use phylogeographic differences and variation in life history characteristics between Pisaster ochraceus and *Pisaster brevispinus*, to test among selective processes that may be important for bindin evolution in this genus. For both species, we find extensive allele sharing at the bindin locus among populations and weak population divergence, comparable to population genetic differentiation measured at the mitochondrial locus, COI. Phylogeny-based tests for positive selection indicate high rates of amino acid substitutions in bindin within each species, notably among those lineages leading to relatively rare and private alleles. However, this strong selection has not caused population-specific adaptation; similar levels of positive selection are found in geographic regions where *P. ochraceus* populations are sympatric and allopatric with *P. brevispinus*. Our results suggest that reinforcement is not an important source of selection for this species pair. The patterns are more consistent with negative frequency dependence and selection for rare bindin alleles, suggesting that sexual conflict may be a common evolutionary process underlying diversification and divergence among *Pisaster* species.

WAS DARWIN RIGHT? POLLINATORS, FLORAL TRAITS AND HETEROSTYLY, AN OLD QUESTION REVISITED IN THE LIGHT OF COMPARATIVE EVIDENCE

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The increasing availability of phylogenetic reconstructions presents the opportunity to test relevant hypotheses about the origin and diversity of plant sexual polymorphisms. Heterostyly, a pervasive discrete floral polymorphism across lineages of flowering plants, has attracted the interest of evolutionary biologists since Darwin's book The different forms of flowers.... Early evolutionary models based on genetics explained the origin and maintenance of heterostyly as an anti-selfing mechanism. The genetic hypothesis contrasted with Darwinian views, which considered heterostyly as a device to promote outcrossing in hermaphroditic plants without the constraints imposed by selfinterference between male and female functions and lack of precision in pollen transfer in hermaphroditic flowers. The mostly ecological Darwinian hypothesis posits a key role for particular pollinators, which are able to efficiently transfer pollen between different morphs. Both hypotheses agree on the fact that heterostyly is a typical mechanism for disassortative mating and is governed by negative frequency-dependent selection, although the factors proposed as important for the evolution of the trait differ. We surveyed occurrence of heterostyly across angiosperms families and genera to trace its evolution across the phylogeny. We determined the number of independent evolutionary transitions from monomorphism to heterostyly, and also the ancestral condition of heterostyly (approach herkogamy or non-herkogamy as suggested by the two competing evolutionary models). Finally we analysed whether the presence of heterostyly is evolutionarily correlated with flower architecture and developmental constraints, such as (1) tubular flowers which restrict pollinator movements and ensure more exact pollen deposition on their bodies, (2) regular flowers which do not restrict pollinator movements within the flower, and (3) flower with free stamens (not connate to the perianth).

30. Phylogenetics and Phylogeography

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DETECTING RECENT CHANGES IN THE DEMOGRAPHIC PARAMETERS OF DROSOPHILID POPULATIONS FROM WESTERN AND CENTRAL AFRICA

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Previous genetic studies showing evidence of past demographic changes in African drosophilids suggested these populations had strongly responded to Quaternary climate changes. We surveyed nine species of *Zaprionus*, a drosophilid genus mostly present in Africa, in forests located between southern Senegal and Gabon. The mitochondrial COI gene showed contrasted levels of sequence variation across species. Populations of the only cosmopolitan species of the genus, *Z. indianus*, and of its closely-related sibling species, *Z. africanus*, are highly polymorphic and appear to have undergone a continuous population expansion beginning about 130,000 years ago. Five less variable species probably underwent a population expansion beginning only about 20,000-30,000 years ago. One of them, *Z. taronus*, was significantly structured between forest blocks. The last two species were nearly monomorphic, probably due to infection by *Wolbachia*. These results are similar to those obtained in three species from the melanogaster subgroup. They may be typical of the responses of African drosophilid populations to glacial cycles.

PHYLOGEOGRAPHY AND POPULATION GENETIC STRUCTURE OF AN ALIEN LIZARD

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Invasive species have been long studied for their ecological and economic impact on the native species. Recently their potential as study systems in evolutionary biology has been recognised since they present an opportunity to study ecological and evolutionary processes over contemporary timescales. The common European wall lizard, *Podarcis muralis*, covers a wide range in western and southern Europe with several genetically and geographically distinct populations. The species was introduced, through multiple introductions, in the UK where it has established more than 25 populations. Identifying the phylogeographic origin of introduced populations is an important first task and we have resolved this using a phylogenetic tree approach. We sequenced a 655bp region of the mtDNA cytB gene and combined unique haplotypes from 500 individuals from 23 non native populations with published sequences from the native range. Furthermore, we genotyped 960 individuals from both introduced and native populations at 16 microsatellite loci to infer their genetic diversity and how is affected by their origin and admixture. We identified 12 unique haplotypes in the introduced populations that were nested into five, geographically distinct, native clades. Multiple clade origin was common within populations, with a maximum of three different haplotype clades being represented within a single population. These results show that human introductions have created substantial opportunities for hybridization between genetically and phenotypically distinct lineages. Overall, the genetic diversity was significantly lower in the non-native populations but mixed-origin populations had higher genetic diversity than single origin populations. Changes in the genetic diversity may have important consequences for the establishment success and long-term viability of introduced wall lizard populations.

EUROPEAN BROOK AND RIVER LAMPREYS, SPECIES OR ECOTYPES: NEW INSIGHTS USING MICROSATELLITE LOCI

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The ice ages are known to be the most dominant palaeoclimatic feature occurring on Earth, producing severe climatic oscillations. The Pleistocene ice ages, together with recent processes, considerably shaped the distribution and population structure of several species. Lampreys constitute excellent models to study the colonization of freshwater systems, as they commonly appear in pairs of closely related species of anadromous versus freshwater resident adults, thus having the ability to colonize unexplored habitats, through the anadromous species, and establish freshwater resident derivates. We used microsatellite loci to investigate the spatial structure, patterns of gene flow and migration routes of Lampetra populations in Europe. Lampetra in Europe is represented by the migratory L. fluviatilis and four resident species, L. planeri, L. alavariensis, L. auremensis and L. lusitanica, the last three endemic to the Iberian Peninsula. We found that in this southern glacial refugium almost all sampled populations represent a distinct genetic cluster, showing high levels of allopatric differentiation, reflecting long periods of isolation. The more recently colonized populations from central and northern Europe are less divergent among them, as result of their more recent common ancestor. They are represented by fewer genetic clusters and lower diversity, and there is evidence of strong recent gene flow among the migratory populations. Finally, we found no signal of hybridization between sympatric populations of *L. fluviatilis* and *L. planeri* from the Iberian Peninsula, and population structure analysis revealed that they constitute two distinct genetic clusters, indicating that these species constitute two distinct taxa that diverged very recently, as previous results from genomic analysis.

DIVERSIFICATION AND GEOGRAPHIC PROGRESSION IN POPULUS (SALICACEAE)

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The flowering plant genus *Populus* comprises ~30 species of deciduous, dioecious trees. Despite the economic, ecologic, and scientific importance of the genus, understanding of patterns of diversification within *Populus* remains poor. The difficulty in resolving relationships is attributable to widespread interspecific gene flow, recent and rapid diversification, and the previous application of a small number of insufficiently variable sequence markers. Using the published poplar genome to develop a large number of sequence markers, we generated and analyzed data from the chloroplast, mitochondrial, and nuclear genomes to estimate sectional and interspecific relationships, dates of divergence for major clades, lineage-specific substitution rates, patterns of geographic progression, and diversification rates within *Populus*.

REVEALING THE ORIGIN OF AN INVASIVE INSECT USING ANCIENT DNA AND HISTORICAL HERBARIA

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Determining the origin of alien invasive species is crucial to developing invasive species management strategies. However, the origin of many alien species remains uncertain because of the lack of historical data. For instance, the moth Cameraria ohridella (Gracillariidae) was described in 1986, as a genus new to Europe and had managed to invade almost all Europe since 1989. Its larvae are leaf miners on the white flowering horse-chestnut (Aesculus hippocastanum), causing significant damage to their summer foliage. The fact that the appearance of C. ohridella in much of Western Europe has been so recent and dramatic, without earlier detection by entomologists, has made its origin a subject of debate . Originally thought to be a relict species in the Balkans, a more recent hypothesis is that the moth is an example of a sudden host plant shift to horse-chestnut, probably from maple or sycamore (Acer spp.), maybe combined with long distance translocation. Examination of horse-chestnut samples in seven historic herbarium collections revealed that almost half of 71 sheets had leaf mines with larvae/pupae inside. This material came from natural populations in Albania and Greece and dated from 1981 back to 1879. We extracted DNA from 54 archival larvae and used five COI minibarcode primer pairs developed specifically for C. ohridella. We successfully amplified DNA minibarcode fragments from 10 larvae extracted from herbarium specimens from 1936 to 1981. These archival sequences confirm an identity and Balkan origin for C. ohridella and the herbarium data set its history back by over a century. The herbaria reveal three previously unknown mitochondrial haplotypes. We also detected local outbreaks back to 1961 and dynamic frequency changes, which may be associated with road development. This case history demonstrates that herbaria are greatly underutilised in studies of invasive species origins, herbivore biodiversity and insect-plant interactions.

PLEISTOCENE SPECIATION AND BIOGEOGRAPHIC HISTORY OF ARCTIC-ALPINE PLANTS

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The climatic change in Pleistocene has drastically influenced on global biological diversity. Due to the beginning of cool climate, species adapted to cold environment obtained the chance to expand widespread range, especially in the boreal region in the northern hemisphere. Thereafter, repeated climatic oscillations caused geographic isolation and genetic differentiation throughout species' range, leading to allopatric speciation. In contrast, repeated changes of species' range following climatic oscillations may allow genetic admixture among diverging lineages and prevent speciation. Here, we examined evolutionary impact of Pleistocene climatic changes by focusing on arctic-alpine plants in genus Phyllodoce (Ericaceae). Molecular phylogenetic analyses based on multilocus sequences of nuclear genes revealed that a monophyly of four boreal species with basal divergence of an alpine endemic one in the Japanese archipelago. Model-base analyses elucidated that the divergence of four boreal species occurred with introgression, even if discarding an apparent signature of genetic mixture beyond species. By calculating divergence time based on general substitution rate, the divergence of four boreal species from Japanese endemic one as well as their diversification were originated in the late Pleistocene (ca. 60-80 yBP and ca. 30 yBP, respectively), during which the climatic oscillations started to be intensified. Consequently, our study demonstrated that diversification of arctic-alpine species certainly occurred following the Pleistocene climatic oscillations regardless of the potential genetic admixture, whereas a marginal endemic in Eastern Asia was a relict species of ancient divergence.

NUCLEOTIDE VARIATION IN THE *PLANTAGO* MITOCHONDRIAL GENOME

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Plantago is a model genus of plant mating system evolution. In addition to great variation in selfing and outcrossing syndromes, many species also exhibit gynodioecy: the presence of hermaphroditic and female individuals in the population. The complex interplay between cytoplasmic sterility factors and nuclear restorer genes that composes the genetic underpinnings of gynodioecy in *Plantago* may contribute to the marked, relative increase in mitochondrial nuclear substitution rate that has been observed. We sequenced 20 mitochondrial genes from multiple gynodioecious and monecious species to determine basic patterns of nucleotide variation within the genus as well as to test the hypothesis that gynodioecy contributes to increased substitution rates in the mitochondrial genome.

DEMOGRAPHIC INFERENCE OF PAST VARIATIONS IN POPULATION SIZE : WHAT IS THE GAIN OF COMBINING DIFFERENT GENETIC MARKERS SUCH AS DNA SEQUENCES, SNPS AND MICROSATELLITES

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Stephens and Donnelly(2000) introduced an efficient importance sampling scheme for computing the likelihood surface of a genetic sample in coalescent-based models. This technique was further extended by De Iorio and Griffiths (2004a,b) for more general population frameworks and mutation models. For DNA sequence loci, the results of De Iorio and Griffiths (DIG) represent an improvement in terms of efficiency over those of Bahlo and Griffiths (2000) but haven't been tested beyond a single panmictic population. In this work, we implemented this approach and derived demographic inferences when modeling variation of the past size of a single population. Our method can analyse microsatellite loci evolving under a generalized stepwise mutation model, DNA sequences evolving under the infinitelymany-sites model and bi-allelic single nucleotide polymorphisms (SNPs). Our method can also simultaneously analyse different types of markers. Using simulations, we tested how the number and types of markers considered, as well as combinations of different markers might influence the precision of demographic estimations. For those tests, we considered two typical demographic scenarios : (1) an old contraction followed by an expansion with large present population size, corresponding to typical post-glaciation recolonization scenarios; and (2) a recent strong contraction with small present population sizes, corresponding to typical scenarios studied in conservation biology. References: Stephens, M., Donnelly, P., 2000. Inference in molecular population genetics. J. Roy. Statist. Soc. B 62, 605–655. Bahlo, M., Griffiths, R.C., 2000. Inference from gene trees in a subdivided population. Theor. Popn. Biol. 57, 79-95. De Iorio, M., Griffiths, R.C., 2004a. Importance sampling on coalescent histories, I. Adv. Appl. Probab. 36, 417–433. De Iorio, M., Griffiths, R.C., 2004b. Importance sampling on coalescent histories, II. Subdivided population models. Adv. Appl. Probab. 36, 434-454

TESTING ENVIRONMENTAL AND GENETIC EFFECTS IN CORRELATED LANDSCAPES

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Correlated random effects are a well-recognized concern for observational data in general, and more specifically for spatial data in ecology. Generalized linear mixed models (GLMMs) with correlated random effects are a potential framework for handling these correlations. However, as the result of statistical and practical issues, GLMMs have been put into practice through of undocumented use of procedures based on penalized quasi-likelihood approximations (PQL), and under restrictive models of spatial correlation. Alternatively, they are often neglected in favor of simpler but more questionable approaches such as partial Mantel tests. In this work we aim to provide practical and validated means of inference under spatial GLMMs, that overcome these limitations. For this purpose, an R package is developed to fit spatial GLMMs. We assess the performance of likelihood ratio tests for fixed effects under spatial autocorrelation, based on Laplace or PQL approximations of the likelihood. Expectedly, the Laplace approximation performs generally slightly better than PQL, although a variant of PQL was better in the binary case, where it could be expected from previous works to perform worst. Finally, we illustrate the efficiency of a bootstrap procedure for correcting the small sample bias of the tests.

CONNECTIVITY OF THE CORAL POCILLOPORA EYDOUXI IN THE SOUTHWESTERN INDIAN OCEAN: A TAXONOMIC CHALLENGE

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Coral reefs are threatened by human impacts and global climate changes. Reef resilience after disturbance depends largely on dispersal and colonisation abilities of corals. We focused on a major constituent of the Western Indian Ocean reefs, the coral *Pocillopora eydouxi*. This branching species is abundant and distributed all around the Indo-Pacific Ocean, constituting a key species for the ecosystem. The aim of this study was to assess the connectivity of 9 populations (n=340 colonies of P. eydouxi) sampled from the outer reef slopes of 3 islands (3 populations per island) in the Southwestern Indian Ocean (Reunion to the south-east of Madagascar, Europa to the South of the Mozambique Channel and Tromelin to the north-east of Madagascar) using 9 microsatellite loci presenting no null alleles nor linkage disequilibrium. Distance between populations pairs ranged from 2 to 1600 km. All the 9 populations presented high significant heterozygote deficiency (FIS = 0.247-0.415 < strong >). Between all pairs of populations, few differentiations were observed (FST = 0-0.07). Only Tromelin Island is differentiated from the others (mean FST*=0.037), but no isolation by distance pattern was found. Nearly all the genetic variance was distributed within populations among islands. Assignment tests using Bayesian statistics showed that, among all individuals of P. eydouxi, four distinct genetic clusters were identified. These four clusters were highly genetically differentiated (FST $= 0.129-0.236^{**}$) but presented no congruence with the geographic origin of the individuals. Thus, these genetic clusters could be linked to the presence of cryptic species within a morphological type or to phenomenon of hybridization between several species. All these results question the taxonomy of the genus Pocillopora based on morphological characters. Nevertheless, the P. eydouxi* populations are highly connected in the Southwestern Indian Ocean.

30. Phylogenetics and Phylogeography

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DISPERSAL AND SURVIVAL IN PACIFIC RIM PEAT MOSSES (SPHAGNUM)

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Mosses mostly exhibit broader distribution ranges than higher plants, coupled with limited morphological differentiation across large distances. This phenomenon has traditionally has been explained by slow evolution in mosses. It has recently been demonstrated that long-distance dispersal can be very high in transatlantic peat mosses (*Sphagnum*), and we ask whether transpacific peat mosses display similar high gene flow rates through time. We also ask where these stress-tolerant plants likely have survived climatic oscillations through Pleistocene.

INTER AND INTRA ISLAND INTROGRESSION IN A WOLF SPIDER RADIATION FROM THE GALÁPAGOS, AND ITS IMPLICATIONS FOR PARALLEL EVOLUTION

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Parallel radiations within island systems are often assumed to follow a simple scenario in which single colonization events are followed by in-situ divergence. However, subsequent gene exchange after the initial colonization and during the divergence process might have evolutionary impacts on species radiations. First, gene exchange among ecologically similar species from different islands may lead to introgression of adaptive genetic variation and, hence, influence the parallel divergence process. Second, hybridization events among ecologically different species living on the same island may influence their genetic relationships due to the introgression of neutral genetic variation. In this study, we estimate levels of gene exchange within a wolf spider radiation of the genus Hogna from the Galapagos, wherein habitat specialization into 'high elevation' and 'coastal dry' species apparently evolved repeatedly on two islands. By using a multilocus approach we show that low levels of inter island and relatively higher levels of intra island introgression have most likely been prevalent within this spider radiation. Using these estimates, we demonstrate by means of a coalescence simulation that adaptive gene genealogies, expected to better reflect the adaptive divergence process, may strongly differ from gene genealogies of neutral genes. As species phylogenies within radiations are frequently used to infer the divergence pattern, interspecific gene flow should not be neglected when interpreting parallel trait evolution.

MTDNA EVOLUTION AND BIOGEOGRAPHY OF THE GENUS PINUS

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Reticulate evolution resulting from interspecific hybridization played a major role in the evolution of plant species. In the genus *Pinus*, mitochondrial (mt) genome is maternally inherited and chloroplast (cp) genome paternally inherited, which provides a unique opportunity to validate ambiguous instances of reticular evolution in this genus. In this study, we sequenced more than 10 kbp mt segments in 31 pine species to characterize the patterns of mtDNA evolution in and infer phylogeographical history of the genus. We found complex patterns of repetitive sequences scattered over the different regions of mt genome, and chimeric structures likely generated by complex recombination events. There were discordance between mtDNA and the previously published cpDNA phylogeny of Pinus, which are hypothesized to have been caused by at least four mtDNA capture events during range expansions of the pine species. These results indicates that both vertical and horizontal inheritance have been involved in the evolution of mtDNA in the genus *Pinus*. By using an 85 MY calibration as the divergence time of the two subgenera *Pinus* and *Strobus*, most of the current lineages were diverged within 20 MYA in both subgenera. However, the isolation history during Eocene and the processes of colonization during Oligocene-Miocene were different between the two subgenera. Subgenus Pinus was preserved in a single Eocene refugium located in a circumpolar high-latitude zone, and subsequently differentiated during Oligocene-Miocene expansion. In contrast, the genetic divergence of subgenus *Strobus* was initiated and reinforced during Eocene. The current distribution pattern of this subgenus was generated by Oligocene-Miocene expansion from three Eocene refugia via both withinand inter-continental dispersals. Our results revealed instances of ancient reticulate evolution in the genus *Pinus*, and shed light on the Eocene impacts on pine distribution and evolution.

ACCOMMODATING HYBRID/INTROGRESSED LINEAGES IN THE MULTISPECIES COALESCENT: THE COMPLEX EVOLUTIONARY HISTORY OF ANTIRRHINUM UNRAVELED

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Antirrhinum L. (snapdragons; c. 25 spp.) is an Old World plant genus primarily distributed in the western Mediterranean basin. Antirrhinum exhibits many interesting ecological and morphological features. Active research has been undertaken on pollination biology and biogeography and some species (mainly Antirrhinum majus L.) have been traditionally much appreciated due to its ornamental value as well as to its importance as a model system for molecular floral development. In spite of this research attention, and regardless the extensive efforts made, the phylogenetic relationships within the genus are far from being well resolved. Previous phylogenetic analyses failed to infer species relationships within the genus. The difficulty in defining species boundaries and the low phylogenetic resolution suggested an early rapid diversification of the genus. Under this scenario, processes obscuring phylogenetic reconstruction such as hybridization/introgression and/or incomplete lineage sorting are likely to have occurred. Moreover, hybridization processes in field and under controlled conditions were previously reported in several Antirrhinum species. In order to obtain accurate phylogenetic relationships within Antirrhinum we obtained allelic data from two low-copy nuclear genes and three plastid loci. We carried out coalescent-based simulations to distinguish hybridization from coalescent stochasticity. Additionally, we incorporated hybrids/introgressed species in the multispecies coalescent model by using a multilabelling approach in which a lineage of hybrid origin is represented with two sources of parental contribution. This methodology allowed the accurate reconstruction of complex evolutionary patterns that have occurred in this group of species extensively used for evolutionary research.

SYSTEMATICS IN PRESENCE OF RETICULATION AND INCOMPLETE LINEAGE SORTING: THE CASE OF *LINARIA* SECT. *SUPINAE*

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We examined the phylogenetic history of *Linaria* with special emphasis on the Mediterranean sect. *Supinae* (44 species). We revealed extensive highly supported incongruence among two nuclear (ITS, AGT1) and two plastid regions (*rpl32-trnLUAG*, *trnS-trnG*). Coalescent simulations, a hybrid detection test and species tree inference in *BEAST revealed that incomplete lineage sorting and hybridization may both be responsible for the incongruent pattern observed. Additionally, we present a multilabelled BEAST species tree as an alternative approach that allows the possibility of observing multiple placements in the species tree for the same taxa. That permitted the incorporation of processes such as hybridization within the tree while not violating the assumptions of the *BEAST model. This methodology is presented as a functional tool to disclose the evolutionary history of species complexes that have experienced both hybridization and incomplete lineage sorting. The drastic climatic events that have occurred in the Mediterranean since the late Miocene, including the Quaternary-type climatic oscillations, may have made both processes highly recurrent in the Mediterranean flora.*

THE PHYLOGEOGRAPHY OF THE COMMON HAMSTER, THE SPECIES ENDANGERED IN EUROPE

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The common hamster (*Cricetus cricetus* L.) is a formerly steppe species, in Europe generally restricted to agricultural sites. The changes in agriculture practice, urbanization of formerly agricultural areas, direct persecution and, most probably, climate change caused the breakdown of populations in many European countries. For any conservation plans, it is essential to gain information about the phylogeographic lineages and their source, refugial populations. In Europe, three phylogeographic lineages were described so far: North in Western Europe and Germany, Pannonia in the Carpathian Basin and E1 lineage in Eastern Poland and Western Ukraine. Paleontological data suggest that the common hamster survived the maximal glaciations in the Ukrainian and European Russian belt of steppes. The aim of this work was to describe the genetic differentiation of the Ukrainian populations, which could serve as a source of westward expansion of hamsters into Central and Western Europe. The phylogeographic analysis was performed on the basis of three sequences of mitochondrial DNA (*the control region, 16S rRNA and cytochrome b*) and six conserved intron loci located on the Y chromosome (*YCATS*). The phylogeographic relationships of the analyzed populations will be described on the basis of the gene trees and networks of haplotypes. The levels of diversity will be compared with other described populations.

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EVOLUTIONNARY DYNAMICS IN THE SOUTHWEST INDIAN OCEAN MARINE BIODIVERSITY HOTSPOT: A PERSPECTIVE FROM THE ROCKY SHORE GASTROPOD GENUS *NERITA*

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The southwestern Indian Ocean (SWIO) is a striking biodiversity hotspot. It hosts the second highest diversity of reef corals and associated reef biodiversity, only slighly below that of the much larger Indo-Malayan diversity hotspot, but with many marine endemics. The island archipelagoes with their diverse marine habitats constitute a natural laboratory for studying diversification processes. Rocky shores have been little studied in the SWIO region. This habitat contains high mollusks diversity, in particular gastropods. To explore the role of climatic and geologic factors in lineage diversification within the genus Nerita, we constructed a new phylogeny with an associated chronogram from two mitochondrial genes (cytochrome oxidase sub-unit 1 (CO1) and 16S rRNA (16S)), combining previously published and new data from samples collected throughout the region. Our results suggest that the regional diversification of Nerita is closely linked to tectonic events in the SWIO region. The Réunion mantle plume has been stable and active for >200 Ma, generating island archipelagoes, partly below sea-level today. Since the Miocene, sea-level fluctuations have intermittently exposed rocky shore habitats. These represent ephemeral stepping stones for intertidal gastropods, which have likely facilitated repeated colonization by Nerita populations from the Indo-Malayan region, leading to allopatric speciation. This highlights the importance of taking into account past climatic and geologic factors when studying diversification of highly dispersive tropical marine species. It also underlines the unique history of the SWIO region, harboring a unique marine biodiversity.

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SPECIATION IN A LARGE TROPICAL AMPHIBIAN RADIATION

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The native amphibian fauna of Madagascar is constituted by five endemic evolutionary lineages of frogs with nearly 100% species-level endemism. Those include the endemic family Mantellidae that shows a wide species diversity within the island. Two undescribed mantellid species were also known from the oceanic Comoros Islands, which were colonized twice by oceanic dispersal ca. 6.5 mya. The recent discovery of a third species of mantellid frog in Comoros, sister and syntopic to one of the known Comoroan species, suggest that they have evolved in sympatry within the island. We here present phylogenetic, morphological, ecological and bioacoustic data of these species and discuss them within speciation scenarios that have been proposed for Mantellid frogs.

HABITAT USE AND SEXUAL DIMORPHISM IN PODARCIS WALL LIZARDS: MICRO- AND MACROEVOLUTIONARY PATTERNS OF MORPHOLOGICAL VARIATION

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Understanding how phenotypic diversity evolves is a major objective of evolutionary biology. Iberian and North African Podarcis are a lizard group of remarkable morphological diversity, thus providing an intriguing model system for studying the causes underlying phenotypic evolution. We examined body size and shape variation at the individual and evolutionary lineage level in relation to sexual dimorphism (SD) and habitat use, to trace the potential effects of sexual and natural selection on different morphological traits. The results obtained indicate that SD is a major source of phenotypic variation in this group, with significant effects on all examined variables at the individual level. Habitat use also explains large part of the variation observed at the individual level, but surprisingly is not associated to body size and limb length variation, as has been the case in other lizard groups. Comparative analyses of trait evolution using the most recent mtDNA phylogeny of the group indicate a significant difference between habitats in male head shape, where males of saxicolous species exhibit relatively flatter and shorter heads than ground-dwelling ones. Such variation is also reflected on macroevolutionary patterns of sexual dimorphism, which also varies across habitats, possibly indicating an interaction between sexual and natural selection. Comparison of evolutionary rates between habitats showed that male head shape has evolved about 9 times faster in saxicolous species. Remarkably, limb length was not associated to habitat use at the macroevolutionary level, contradicting predictions of the ecomorphological paradigm and previous observations on other lizard groups. Indeed, a comparison of evolutionary rates across traits demonstrated that head dimensions have evolved much faster in this group of lizards as compared to limb length. Together our results shed new light to the evolution of the remarkable morphological diversity of this lizard group.

POPULATION GENETIC AND PHYLOGEOGRAPHIC STRUCTURE OF HADAL AMPHIPODS

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The hadal zone comprises ocean depths below 6000m and represents the deepest 45% of the oceans. It consists of trenches that are formed along subduction zones and are characterised by unique topography, geographical isolation, sporadic food availability, low temperatures, high pressures and the absence of natural light. Hadal trenches are relatively unexplored and are thus one of the least understood habitats on the planet. From a biogeographical perspective, hadal trenches have been considered as isolated "islands" with little genetic exchange between trenches and high levels of species endemism. Such as assertion is difficult to reconcile, however, given some species appear to have a cosmopolitan distribution across the abyssal and hadal zones. Such contradictions highlight how little is known about hadal ecology in general and modes of dispersal, levels of gene flow and drivers of phylogeographic structure in particular. Here we examine patterns of genetic divergence between trench populations of two Lysianassoidea amphipod species (Hirondellea dubia and Paralicella tenuipes) which constitute a significant and ubiquitous component of the hadal fauna and are key members within hadal food webs. A suite of species-specific microsatellite markers were developed and used to examine levels of population genetic divergence among populations both within and between trenches around the Pacific Rim, where 75% of hadal trenches are found. This data is combined with mito-genome DNA sequence polymorphisms to resolve overall levels of phylogeographic and population genetic structure and using a landscape genomics approach to assess different hypothesized routes of gene flow and ancestral colonisation.

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RECONSTRUCTING THE ORIGIN AND SPREAD OF YELLOW-NECKED MOUSE APODEMUS FLAVICOLLIS (RODENTIA, MURIDAE) IN THE BALKANS

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The reconstruction of the glacial and postglacial history of species has been in a focus of many genetic researches. A comparative analysis of the levels and patterns of genetic variation across a range of Palaearctic animal taxa has revealed the existence of refugia in the Iberian, Italian and the Balkan peninsula. Numerous studies described the Serbia as a meeting point of different recolonization routes of *A. flavicollis*. The aim of this study is to assess the population divergence and effects of present and past microevolutionary processes on genetic differentiation of populations. Due to the existence of different natural barriers between populations, we expected to find different genetic lineages that survived last glaciations in independent local Balkan refugia. By using AFLPs, we found evidence for significant differences in genetic diversity and genetic differentiation between four populations that are relatively close in space, but separated by major barriers such as rivers. The analyzed populations formed three clusters, corresponding to the three genetic lineages that survived last glaciations in the Balkan refugia where they diverged separately. In silico data sets of genetic polymorphism under specified scenarios were generated by forwards-in-time and backwards-in-time simulators in order to describe the evolutionary history and genetic architecture of a species. The comparison of simulated data and real samples suggests that population divergence in A. flavicollis can be best explained through complicated migration scheme that includes spatial expansion followed by island model of population structure. The observed patterns are reviewed in terms of defining phylogeographic relationships among extant populations. In order to better understand the evolutionary history of the other species in Balkan refugia, divergence time estimates and the effects of geographic barriers to gene flow in general are going to be discussed

PISUM GENOME VARIABILITY DETECTED BY AFLP AND RGA PROFILING

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Pea (P. sativum L.) is an important legume grown and consumed extensively worldwide. In the present study for the first time two different multiloci techniques - AFLP and RGA-profiling have been used for genetic diversity assessment and taxa boundaries delimitation in Pisum species. RGA-profiling technique was used to determine NBS-LRR plant pathogen resistance genes polymorphism. In contrast, AFLP technique allows to investigate mostly evolutionary neutral genome sequences. Both approaches can provide comprehensive genome diversity information. Genetic diversity of 110 Pisum accessions of different origin representing P. fulvum and P. sativum species, including abyssinicum, asiaticum, transcaucasicum and arvense accessions with unclear taxonomic status was analyzed using AFLP and RGA- profiling. By using preselected primer combinations 405 AFLP and 171 RGA polymorphic bands have been detected. As a result, for each pea genotype unique AFLP and RGA-specific patterns have been revealed. The dendrogram and PCA based on calculated GD values revealed two distinct groups corresponding to P. sativum and P. fulvum. Other taxa formed one mixed group with P. sativum accessions. Notably, on AFLP, PCA-plot all P. sativum formed continuum with two main poles, largely represented Asian or European accessions. On RGA-profiling PCA-plot, no geographical and taxonomical differentiations within P.sativum accessions were determined. No correlation between pea accession grouping and resistance status or ability to interact with arbuscular mycorrhiza was found. Obtained data suggested that genus Pisum is composed only of two clear species: P. sativum and P. fulvum. There is no differentiation between abyssinicum, asiaticum, arvense, transcaucasicum and sativum, that confirm their subspecies taxonomic status. Molecular characterization of Pisum genetic diversity levels may help to identify potential new sources useful in pea breeding. The work was supported by FRP "Biological Diversity".

ARE THE DIVERGENT MIGRATION DESTINATIONS OF BOGONG MOTHS AGROTIS INFUSA (NOCTUIDAE: LEPIDOPTERA) RELATED TO GENETIC DIFFERENTIATION?

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Long-distance seasonal migration mechanisms are still unclear, albeit their importance to species persistence and ecosystem functioning. Many populations have well defined migratory destinations and this might have important consequences on their genetic structuring: whereas different breeding grounds and divergent migratory pathways and destinations may result in a lower degree of genetic mixing hence promoting genetic differentiation, the use of common breeding and overwintering/summering grounds may result in a lack of genetic differentiation. Every spring, adult Australian Bogong moths (Agrotis infusa) migrate from western New South Wales and south-eastern Queensland to the Snowy Mountains (the highest peaks of the Great Dividing Range) to estivate in cool rock crevices and caves; they then return to their breeding grounds during the autumn to mate. During estivation, the moths are gregarious and remain in diapause serving as food for many insectivorous animals given their high body fat content (around 65%). Owing to their different summering sites, some may fly up to 1000 km in a single direction. Although some attempts to use genetic markers to identify specific natal sites of adult Bogong moths have been made, it is still unknown whether the different migratory pathways and destinations have a genetic basis. In summer 2012-2013, Bogong moths were collected live by hand at four estivation sites in New South Wales (Mount Gingera, South Ramshead, Mount Morgan, and Mount Tingaringy) and examined for sequence and haplotype diversity and genetic distance at mitochondrial and nuclear genes. The data will elucidate if differences in the migration routes and summering sites of these moths depend on—or have resulted in—genetic divergence, which will be essential for Bogong moth conservation. The Bogong moth is an iconic Australian species and the protection of their summering sites may be crucial to maintain species variability.

HYBRIDIZATION IN A RECENTLY REDISCOVERED DOLPHIN SPECIES, STENELLA CLYMENE

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Hybridization has long been considered to play a major role in the evolution of plant and animal taxa, occurring as a simple exchange of interspecific genetic material, or even originating new species. In mammals, reports of hybridization and hybrid speciation are particularly rare when compared to other taxa like insects and fish, which is either a consequence of unfavorable ecological and physiological conditions, or a lack of empirical studies. In cetaceans, there have been several reports of hybridization in captivity and a few others in the wild. The prominent karvological uniformity found to exist in these species may justify the potential to produce viable hybrid offspring more easily than other mammals. In this study we use sequences from one mitochondrial and six nuclear DNA regions in order to understand the evolutionary origin of the Clymene dolphin, Stenella clymene, which is thought to have originated through hybridization among two other species, Stenella coeruleoalba and Stenella *longirostris*. We report a discrepancy of results between markers, with the mitochondrial genome of *S*. *clymene* being more similar to that of *S*. *coeruleoalba*, and the nuclear genome being more similar to *S*. longirostris. We have also found two S. clymene x S. longirostris hybrids, confirmed with genetics and morphological characters. We suggest that introgressive hybridization and hybrid speciation have been important phenomena in the origin of the Clymene dolphin. Overall, our results have strong implications not only for the clarification of the evolutionary relationships among species of the genus Stenella, but also for our understanding of hybrid speciation and the establishment of reproductive isolation barriers in mammals.

HIGH RELATEDNESS AND CHAOTIC GENETIC PATCHINESS OF A POLYCHAETE IN A HETEROGENEOUS ESTUARINE LANDSCAPE

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Population genetic structure in marine organisms frequently defies clear explanation and is described as chaotic genetic patchiness. In these cases, patterns of genetic structure may be determined by barriers to dispersal that are difficult to observe, e.g., those created by variable oceanic currents or by the behavior of dispersive larvae and adults. We examined population genetic structure using seven microsatellite loci in the polychaete worm, *Pygospio elegans*, sampled from 16 sites in a heterogeneous estuarine landscape in Denmark. Most samples showed significant pairwise Fst and Jost's D, and there was no indication of isolation by distance. Using GESTE, we calculated sample-specific Fst values, which characterize how the samples differ from the metapopulation as a whole. Variation in these estimates was not explained by environmental variables, including salinity, substrate, and distance from the mouth of the estuary. The samples most dissimilar in terms of observed allele frequencies, and those with high estimated numbers of siblings, had larger sample-specific Fst. We hypothesize that polymorphism in the developmental mode of *P. elegans*, which produces both dispersive planktonic larvae and non-dispersive larvae brooded by the mother, contributes to chaotic genetic patchiness in this estuarine landscape. The large numbers of siblings estimated within samples could reflect high local recruitment of brooded larvae, despite effective dispersal and high gene flow via planktonic larvae.

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PATTERNS OF GENETIC DIFFERENTIATION IN A WIDESPREAD PREDATOR INSECT SPECIES ASSOCIATED WITH GREENHOUSE CROPS, COENOSIA ATTENUATA (DIPTERA: MUSCIDAE)

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Adults of the tiger-fly, *Coenosia attenuata* Stein, are the only known predators of adult stages of important greenhouse crop pests such as whiteflies and leafminers. *C. attenuata* has been recognized as a potential biological control agent in greenhouses but no studies to date have been done to characterize the genetic variation and differentiation across the distribution range of the species. The phylogeographic study on samples from Europe, North and South America and Western Asia based on cytochrome oxidase I mitochondrial gene reveals generally high haplotype diversity but low nucleotide diversity, suggesting a recent expansion, and shows evidences of a recent colonization of North and South America from the Palearctic. The probable origin of the species within the Paleotropical region are discussed, as well as the dispersal patterns of the species and the dates of divergence of populations.

DRAWING GEOGRAPHIC AND GENETIC BOUNDARIES IN A PLANT BIODIVERSITY HOTSPOT: CRYPTIC NARCISSUS SPECIES FROM THE SOUTHERN IBERIAN BAETIC RANGES

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Species delimitation is a fundamental issue for many fields of biology and it is also a first step towards understanding evolutionary mechanisms generating diversification. Certain regions of the Mediterranean Basin preserve an extraordinarily rich flora in combination with an exceptional concentration of endemic taxa. Because the spatial genetic structure of populations often plays a key role in speciation, studies focusing on the genetic structure of closely related endemic taxa would be valuable to understand the evolutionary forces involved in the diversification processes. The Baetic mountain ranges, a remarkable biodiversity hotspot in the southeastern Iberian Peninsula, harbour several described trumpet daffodils (Narcissus L., section Pseudonarcissi DC.) that are narrow endemics, although considerable disagreement exists among plant taxonomists over the number and rank of taxa involved. We take advantage of amplified fragment length polymorphism (AFLP) technique for delimiting taxa within this complex group of wild daffodils. In particular, we tested whether divergent genetic lineages exist rather than a single, genetically homogeneous entity. To this end, we analyzed AFLP data from a broad geographic sampling of 36 natural populations covering the whole distribution area and encompassing as much ecological and morphological diversity as possible. Results indicate that, despite their remarkable phenotypic homogeneity, trumpet daffodils occurring in the Baetic Ranges are a genetically highly heterogeneous group that can not be ascribed to one single taxon as sometimes proposed. On the contrary, four distinct genetic lineages can be identified within the area, whose distribution bears evident relationships with orographic regional features and ecological features of associated microhabitats. Our results emphasize that assessment of plant species richness by conventional methods may considerably underestimate the conservation value of biodiversity hotspot.

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LOW GENETIC DIVERSITY IN BORNEAN ELEPHANTS (ELEPHAS MAXIMUS BORNEENSIS) CONFIRMED BY HIGH THROUGHPUT SEQUENCING METHODS

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The Bornean elephant is a subspecies of mainland Asian elephant which differs from other subspecies in its morphology and behaviour. Its distribution is restricted to the North of Borneo where the estimated population size is around 2,000. They are classified as endangered according to the IUCN Red list of threatened species. The main threats identified are habitat fragmentation and habitat loss due to oil palm plantations. Modern analytical methods for population genetics inference are expected to provide more accurate results when data from multiple genome-wide loci are analysed. To study the effect of recent fragmentation on genetic diversity of Bornean elephants, we developed panels of genomic markers (microsatellites and SNPs) using two different sequencing platforms, the Roche 454 (shotgun) and Illumina (Restriction site associated DNA, RAD). Approximately, 6,683 (shotgun) and 14,724 (RAD) SNPs were detected within our elephant sequence dataset. We found around 2000 SNPs as potential candidates for genotyping assay. A subset of SNPs were tested against a panel of seven elephant blood samples which resulted in a successful validation rate of 83 to 94% and 17% of the loci were found to be polymorphic with a low diversity (He=0.051, Ho=0.057). We have started to validate

these genomic markers on fecal samples of Bornean elephants from different regions of northern Borneo to study patterns of genetic diversity and differentiation across the species range. Different numbers of microsatellites were also identified through these techniques. All the microsatellites tested were monomorphic in the Bornean samples but were polymorphic in a small sample of another Asian elephant subspecies. Our project is one of the still few that have identified genomic markers in a threatened non-model species and that demonstrate the utility of high throughput sequencing technologies as a starting point for the development of genomic tools for a species with low genetic diversity.

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CRYPTIC AVIAN DIVERSITY IN THE SULAWESI SUB-REGION: A CASE STUDY FROM THE WAKATOBI ISLANDS

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Species are the fundamental unit within evolutionary, biodiversity and conservation research. Accurate knowledge of the number of existing species is required to further our understanding of the patterns and processes that shape biodiversity. Cryptic species - two or more biologically distinct species classified as a single species due to morphological similarity - are therefore of great importance to evolutionary research. The discovery and recognition of such cryptic species requires an integrative, multi-disciplinary approach. We examined the taxonomic status of the Grey-sided Flowerpecker (*Dicaeum celebicum kuehni*) populations endemic to the Wakatobi islands. We present evidence from morphometric, genetic and phylogenetic analyses that describe the Wakatobi populations as a distinct, independently evolving species endemic to the Wakatobi archipelago. As the Wakatobi islands host a further five endemic subspecies, we feel these populations warrant re-examination. Previous studies of cryptic diversity in the Sulawesi sub-region have focussed predominantly on morphological, acoustic and phylogenetic techniques in future studies is essential to maximise the detection of cryptic diversity in this biologically rich sub-region.

ON THE ORIGIN OF THE ENDEMIC PIGEON COLUMBA PALUMBUS AZORICA

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Recent studies have been focusing on the phylogenetic relationships among the endemic *Columba* from the Macaronesia – C. palumbus azorica (Azores Islands), C. trocaz (Madeira Island), C. bollii and C. junoniae (both from Canary Islands) – through several approaches, namely mtDNA and microsatellites, with somewhat incongruent results. In the present study, the origin of the endemic subspecies *C*. *p*. azorica was inferred based on five nuclear fragments. Overall, C. p. azorica shows higher haplotype diversity and nucleotide diversity values than the mainland subspecies C. p. palumbus and the Madeiran *C. trocaz* for the most introns analysed, which was not expected since the insular populations frequently have less genetic diversity than the mainland populations. Our results also support a previous study in which it is described that *C. trocaz* is phylogenetically closer to *C. bollii* than to *C. p.* palumbus (Dourado et al., in press). Surprisingly, for the Azorean subspecies, while some samples cluster with the mainland C. p. palumbus, others share the same group with C. trocaz. The most plausible hypothesis to explain these results is a colonisation of the Azores by an ancestral of *C. trocaz* and *C. bollii*, that then diverged due to geographic isolation. More recently, the mainland subspecies *C*. p. palumbus reached those islands, conducing to admixture between both groups. Hybridization may have conducted to introgression of mtDNA from C. p. palumbus into the other individuals that inhabited Azores Islands, and mtDNA from *C. p. palumbus* prevailed. Therefore, instead of a simple isolated woodpigeon population, *C. p. azorica* is a much more complex and interesting event in pigeon evolution than formerly alleged.

D21SY30PS0999

PHYLOGEOGRAPHIC HISTORY OF THE FLAT PERIWINKLES, LITTORINA FABALIS AND L. OBTUSATA

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The flat periwinkle *Littorina fabalis* is an intertidal gastropod distributed in the NE Atlantic from Iceland and the White Sea in the North to Portugal in the South. It exhibits several ecotypes along its range and it represents a promising system to explore the mechanisms of ecological divergence as well as of parallel evolution. Nonetheless, prior knowledge of the phylogeographic history of the species is required to address these questions. Previous studies suggest a complex diversification process among Northern European *L. fabalis* ecotypes, further complicated by the interaction with its sister species *L. obtusata*. Both species co-occur in most European shores and, while they are well differentiated in terms of ecology, morphology, allozymes and microsatellites, they share substantial mitochondrial variation. With the ultimate goal of understanding to what extent the observed intra and interspecific patterns could result from single or multiple events of adaptation, we perform a comprehensive phylogeographic assessment of *L. fabalis* and *L. obtusata*. We cover the whole distribution range, including all *L. fabalis* ecotypes described so far, and use both mitochondrial and nuclear sequence data to shed light on the demographic history of the flat periwinkles, setting an important baseline to study the above mentioned mechanisms of parallel evolution and ecological speciation.

D21SY30PS1001

PHYLOGEOGRAPHY OF ORTGIESIA SUBGENUS (BROMELIACEAE) AS A CONTRIBUTION TO THE EFFORT IN UNDERSTANDING THE HISTORICAL PROCESSES INVOLVED IN THE SPECIES DIVERSIFICATION IN SOUTHERN PORTION OF THE BRAZILIAN ATLANTIC FOREST

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The number of phylogeographical studies of South America is lower than other regions, despite it having one of the highest levels of biodiversity on Earth. These studies are essential for a better understanding of all the biomes of South America as a whole. Here we studied a group of seven species from *Ortgiesia* subgenus (Bromeliaceae) which are endemic of the southern Brazilian Atlantic Forest (BAF). A relatively few phylogeographic studies were undertaken in this region so far that explain the high number of endemic species and high genetic diversity encountered. To understand the evolutionary history of this group and also of southern BAF, we analyzed the sequences of two plastidial intergenic spacers and one nuclear region from populations throughout the known distribution of all seven species. Thirty-five haplotypes were found for the plastid markers with three of them shared by some species. For the nuclear region, 10 haplotypes were observed and the most frequent one was shared by all the species. This sharing suggests the persistence of ancestral polymorphisms during speciation events and/or hybridization, since individuals with intermediate morphology can be found. In general, species with widespread geographical distribution showed the greatest genetic diversity levels (Aechmea caudata and A. kertesziae). The exception was A. comata that showed high genetic diversity despite it only occurs on an island. High genetic structure was observed for cpDNA; however for the nuclear region, no genetic structure occurs. These preliminary results indicate that some of the species could be affected by the Pleistocene climatic oscillations as already observed for other species of the BAF. More analysis are in progress to confirm this pattern and will be very important in understanding the complex and high levels of biodiversity in South America, and may provide valuable insights into the historical processes underlying diversification in this region.

EVOLUTION OF HAPLOID CHROMOSOME NUMBERS IN THE SUNFLOWER FAMILY

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The number of chromosomes is a remarkably dynamic feature in eukaryotic evolution. In angiosperms, a wide variation in chromosome numbers has already been reported. Considering this large variation, and the ample evidence that drastic genome rearrangements, such as fission, fusion or, more commonly, genome duplication, can contribute to speciation, it is of upmost importance to entangle the evolutionary direction and frequency of such phenomena. Many of the inferences of chromosome number evolution in plants are based on the concept of basic chromosome numbers ('x'). However, a recent paper exploring chromosome numbers evolution in Araceae suggested that, due to this approach, most of studies might be biased towards low chromosomes numbers, producing an inaccurate view of the evolution of chromosome numbers in plants. Also, the use of probabilistic methods of recent models specific for chromosome evolution and of phylogenetic hypotheses might enable to obtain a more robust inference of the evolution of chromosome numbers. In the present study, it was our aim to study the evolution of chromosome numbers in the largest family of flowering plants, Asteraceae. In this family, evolutionary relationships among genera have been established, and a large data set of chromosome numbers is readily available in several web databases. For that, the haploid numbers of all taxa included in the largest phylogenetic hypothesis available for this family (over than 700 genera) were collected, and this data was used to infer the evolution of the haploid number using specific probabilistic models for chromosome numbers. The results of this study will be presented and discussed according with the current trends of chromosome evolution, and will enable to have a better understanding of the processes underlying changes in number of chromosomes, important for understanding speciation and adaptation within this family.

A FIRST GENETIC STUDY OF *MICROCEBUS TAVARATRA*, A LITTLE KNOWN MOUSE LEMUR FROM THE NORTH OF MADAGASCAR

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Madagascar is one of the most important Biodiversity Hotspots in the world. Still, many areas are poorly studied, as is the case of Daraina in the north of the island. In 2010, the highly fragmented forests of this region were surveyed, and the presence of several individuals belonging to the mouse lemur genus (Microcebus) was recorded. In the field, it proved difficult to identify the species encountered by using morphometric measurements, and mitochondrial DNA (mtDNA) was sequenced. Furthermore, seven nuclear loci (microsatellites) were genotyped. Altogether three mtDNA regions (COII, Cyt b and D-loop) were sequenced for 72 Microcebus individuals sampled in three forest fragments (Binara, Solaniampilana and Bekaraoka), in order to determine if all members belonged to the same species (*M. tavaratra*) as two individuals previously identified in this same region by other researchers. We also wanted to describe the patterns of genetic diversity of the sampled populations and determine if genetic clusters could be identified and potentially associated to specific features of the habitat, such large rivers or roads. The results suggest that all individuals belong to the *M. tavaratra* species and that high genetic variation at the nuclear level is maintained in all three populations. The same is observed at the mitochondrial level, with the exception of the population from Binara, which surprisingly did not present any genetic diversity across all mtDNA loci. Moreover, no clear clustering could be defined at the nuclear level, possibly due to the limited number of loci. Despite being currently the most geographically widespread study on Microcebus from Daraina, our results suggest that more research should be carried out across the whole region by increasing both the number of samples and loci. Studies like this one are important for the implementation of appropriate and effective conservation plans for threatened species.

D21SY30PS1021

MITOGENOMICS AND THE PHYLOGENY OF FROGS

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Anurans have a long evolutionary history of more than 200 million years, having undergone an extraordinary diversification that gave rise to more than 6,000 extant species. Previous morphological and molecular studies greatly contributed to the knowledge of the anuran Tree of Life. Yet, important questions remain still under debate. A data set of complete mitochondrial genomes and nine nuclear loci was used to address key questions regarding the higher-level relationships in frogs: (i) the placement of the root in the frog Tree of Life, (ii) the branching order among the five major lineages, (iii) the phylogenetic position of basal relict families within modern frogs (Neobatrachia), and (iv) the internal phylogeny of the family Pipidae (Xenopus' relatives). Our data set demonstrated good phylogenetic performance in resolving the proposed questions, and further allowed examining other issues of great interest, including (vi) the evolution of mitochondrial gene orders in Anura, (vii) the presence of accelerated molecular evolutionary rates associated with the origin of Neobatrachia, and (viii) the evolution of sound production mechanisms in tongueless frogs (family Pipidae), which incorporated new bioacoustic and morphological data.

D21SY30PS1022

EVOLUTIONARY AND BIOGEOGRAPHIC HISTORY OF THE MACARONESIAN BETA-PATELLIFOLIA SPECIES (AMARANTHACEAE): COMBINING PHYLOGENETIC AND ECOLOGICAL STUDIES

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Understanding the origins and diversification of oceanic island flora has long fascinated evolutionary biologists, ever since Darwin. The Macaronesian Islands, in the North-East Atlantic Region, are considered a hot-spot of biodiversity and contain a large proportion of endemic taxa. Recent molecular studies are providing insights into the patterns of colonization and radiation within the endemic plant species, while the relationships between the wild (i.e. endemics and natives) and the crop species have not been suitably addressed from an evolutionary perspective. The aim of this study is to understand the evolutionary and biogeographic history of the seven native species of Macaronesian Beta-Patellifolia species complex (Subfamily Betoideae, Amaranthaceae), which are ancestral species of the cultivated sugarbeet species (Beta vulgaris subsp. vulgaris). We compare the patterns of diversification identified for Macaronesia with those of the Mediterranean Region to assess to what extent patterns may be generalized across regions. Phylogenetic analyses based on nuclear (ITS) and plastid (rbcL, matK, trnH-psbA, trnL-F) DNA markers produced similar topologies that suggest a recent colonization event in the Macaronesian Region. Despite extensive sampling, low levels of sequence variation are consistently found throughout Beta spp., thus preventing complete resolution among taxa. Origins and diversification of these endemics, natives and crop species within the Macaronesian Region will be discussed in a biogeographic context.

PHYLOGEOGRAPHIC STRUCTURE OF THE SPITTLEBUG PHILAENUS SPUMARIUS (HEMIPTERA, APHROPHORIDAE) BASED ON MITOCHONDRIAL DNA AND RAD SEQUENCING

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We investigated the evolutionary history of the meadow spittlebug *Philaenus spumarius*, a widespread and polyphagous insect species of the Holarctic region, limited in its range by the presence of enough humidity in the earlier life stages, and that has a remarkable dorsal colour/pattern polymorphism of the adults. The mitochondrial gene cytochrome c oxidase subunit I (COI) revealed three main haplogroups in Europe: the "western", the "eastern-Mediterranean" and the "north-eastern" haplogroups, which overlapped in some regions. Evidence of recent divergence events at less than one million years ago in southern Mediterranean peninsulas followed by northward population expansions were found. Recent gene-flow events between the main southern peninsulas and between the Iberian Peninsula and North Africa were also detected. A probable British origin for the insular populations of the Azores and New Zealand was revealed, as well as multiple geographic origins for North American populations from western and northern Europe. Restriction site associated DNA (RAD) sequencing was also applied in a set of 8 populations from the different mitochondrial haplogroups and the differentiation patterns obtained from a set of over 15,000 SNPs were concordant with COI results and with a better resolved tree.

D21SY30PS1045

DEEP DIVERGENCE BETWEEN POPULATIONS OF ARANEUS VENATRIX (ARANEAE: ARANEIDAE) FROM BRAZILIAN RAINFORESTS

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Neotropical rainforests (Amazonia and Atlantic) are among the most biodiverse regions of the world, with high endemism and species richness. These predominantly moist and dense biomes, although separated by a large dry corridor of open vegetation, exhibit similar climatic conditions and biotic composition. Geomorphological and paleopalinological data suggest an historical connection between these rainforests, but few phylogeographic studies focus on understanding these biomes formation and species diversification. We investigated the genetic structure and phylogeographic patterns of *Araneus* venatrix (Araneae: Araneidae), a spider with wide geographical range in Neotropics and predominantly found in humid environments. We sequenced mitochondrial COI and nuclear ITS2 regions of populations sampled along the species distribution. Molecular diversity parameters and pairwise FST indicated high genetic variability and strong population structure in this species. Analysis of molecular variance (AMOVA), haplotype networks and phylogenetic inferences revealed a deep divergence between two major groups, separating Atlantic Rainforest populations from the other samples. Molecular clocks with fossil calibration dated this divergence on Late Miocene (7.86 Ma, 95% HPD = 2.7–14.1 Ma). Our results suggest that the evolutionary processes responsible for the differentiation of A. venatrix lineages started before Quaternary, and do not reject hypotheses of Neotropical rainforests' disjunction earlier than Pleistocene.

COAGULATION CASCADE: AN EVOLUTIONARY PERSPECTIVE ACROSS VERTEBRATES

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Blood coagulation is a defense mechanism that enhances organism survival by preventing uncontrolled blood loss in vertebrates. This defense mechanism has to be tightly controlled so the clot forms when and where it is required. Birds exhibit prolonged coagulation time relative to values obtained for mammals. The prolonged coagulation time phenotype observed in birds is shared with reptiles, and within Aves an inter-specific variation has also been described. Despite biochemically and physiologically extensively studied, our understanding about the evolution of the coagulation network in vertebrates is still lagging. Given the increasing availability of genomic-level data we set to uncover the evolutionary story of this fine-tuned network, with a particular emphasis in Aves. The human transcripts for eighteen genes known to encode proteins associated with coagulation response were blasted against the protein coding genes of extant vertebrates available from genomic databases. We included mammals, birds, reptiles, amphibians and fishes. The orthologous proteins were aligned, coverage and protein identity estimated. We found that the clotting cascade in birds involves fewer genes than the mammalian one. The main difference uncovered pertains the membrane receptors from platelet (mammals)/thrombocytes (non-mammals). We propose that the mammalian innovations, i.e. factors only present in mammals, stem for the platelet ability to form resistant clots. And we put forward, that this might be an efficient mechanism for fast and effective control of bleeding from the placental site to prevent fatal hemorrhage during delivery.

PHYLOGENY AND PHYLOGEOGRAPHY OF THE FRESHWATER FISH GENUS SQUALIUS (TELEOSTEI, CYPRINIDAE) IN BALKAN REGION

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The Balkan region is one of the most interesting areas of the Mediterranean slope due to it shares ichthyofauna from two different and isolated regions: independent small/medium size rivers with a Mediterranean fluvial regime, and probably a shared paleogeographic history, and the Danube basin. One of the genera inhabiting this region is *Squalius*, which is widely distributed in Eurasia. Some studies have deal with the phylogenetic relationships of this genus at interspecific level in Balkan Peninsula; however, to date not hard efforts have been done to explore their intraspecific relationships and phylogeography. Besides, new species has been described in the last few years, which phylogenetic position is not known yet. In this study, we investigate the phylogeny and phylogeography of the species of the genus *Squalius* occurring in Balkan region. Thereby, we analyze mitochondrial and nuclear genes to infer the species tree and the phylogeographical structure of Balkan *Squalius* species, the estimated time of their main cladogenetic events and the historical processes that shaped the evolutionary history of this genus in the Balkan region.

D21SY30PS1049

PLIO-PLEISTOCENE EFFECTS ON THE PHYLOGENY, PHYLOGEOGRAPHY AND HISTORICAL EVOLUTION OF FRESHWATER FISH SPECIES LUCIOBARBUS SCLATERI (TELEOSTEI, CYPRINIDAE) IN THE WESTERN MEDITERRANEAN REGION

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The Iberian Peninsula has been traditionally considered as glacial refugia, since glaciers were limited to mountain areas. The concept of refugia is linked to population reduction during cooler periods and expansion once the climate turns back to mild temperatures. In the case of continental icthyofauna, the postglacial spreading is strongly conditioned by the hydrogeographical configuration and could affect the phylogeographic structure throughout time, due not only to its low dispersal power but also to a reduced effective population size. Currently, these refugia are considered heterogeneous and refugia within refugia have been proposed. One of the species used as a model to assess this approach has been the barbel species Luciobarbus sclateri (Günther, 1868), which is an endemism of Iberian Peninsula southern rivers. Thereby, this study aims to know which effects have climatic changes during Plio-Pleitocene on Luciobarbus sclateri, as well as trying to explain, through genetic data, why this species is clearly dominant in those freshwater fish communities where it is distributed. Our hypothesis is that its particular biology and ecological tolerance have resulted to a high genetic variability and scarce population structure. So far, others studies only have dealt with mitochondrial gene, due to the problems related to tetraploidy. Therefore, we analyze mitochondrial and nuclear genes to infer the species tree and its phylogeographical structure, as well as estimate time of the historical processes leading to the evolutionary history of this Iberian endemism.

D21SY30PS1052

PHYLOGEOGRAPHICAL PATTERNS OF NEPHILA CLAVIPES (ARANEAE: NEPHILIDAE) IN NEOTROPICAL RAINFORESTS

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The two great Neotropical rainforests, Amazonian and Atlantic, are separated by a large corridor of drier vegetation, but share many taxa at family, genus and even species levels. There are evidences that the two forests had been connected in the past, and phylogeographical studies on species occurring in both biomes can help to elucidate the evolutionary history of these biomes. *Nephila clavipes* is a widely distributed Neotropical spider species found in these two rainforests and also in some drier areas between them. We sequenced a 491 bp COI mitochondrial region of 323 individuals from 15 sites along central and south regions of Atlantic Forest and 9 individuals from one site in south Amazonia. Our data show the presence of three major mitochondrial lineages, not entirely related to geographic distribution. The samples from Amazonia grouped with some central Atlantic populations on the haplotype network, despite the great geographic distance. Divergences among lineages were estimated on the Pleistocene, between 680 and 510 thousand years ago; data also suggest that the latest expansions events took place around 250 thousand years ago. The climatic fluctuations of Pleistocene may have played a role on the divergence and radiation of the lineages, as also found for vertebrate and plant species. Our study reinforces the role of Pleistocene climate changes in shaping the diversity in the Neotropics.

D21SY30PS1080

PHYLOGENY RECONSTRUCTION IN THE PRESENCE OF INTRA-INDIVIDUAL SITE POLYMORPHISMS: GETTING YOUR SUPPORT BACK

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Intra-*i*ndividual site *p*olymorphisms (2ISPs, e.g. Y or R) in DNA sequences can arise from sequencing errors, or the presence of numerous variants within a multi-copy gene region such as the internal transcribed spacers (ITS) from the nuclear-encoded 35S rDNA cistron. If 2ISPs are due to the latter, and arose from a lack of concerted evolution (and not hybridisation), these sites still offer phylogenetic information. At present, the majority of tree inference algorithms either remove 2ISPs or treat them as ambiguities. This reduces the information content of the dataset and/or creates site uncertainty which in turn can reduce support. Here we propose 2ISP-informative implementations for distance-based, maximum parsimony and maximum likelihood algorithms. These implementations are compared against the standard methods (with 2ISP-ambiguous treatments) using simulated datasets, a meta-analysis from over 20 published datasets, and two case studies. We show that the 2ISP-informative approach offers improved phylogenetic resolution and support as the 2ISP content increases in the dataset. Importantly, hybrid samples with 2ISPs do not result in falsely inflated branch supports under the informative approach. We envisage that this informative approach will greatly aid phylogenetic inference using any nuclear DNA regions that contain 2ISPs (including consensus sequences generated from next generation sequencing), especially at the intra-generic or intra-specific level.

D21SY30PS1092

FINE-SCALE SPATIAL GENETIC STRUCTURE OF A PROTECTED FOREST INSECT

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Understanding the spatial genetic structure of populations can provide insight into ecological and/or evolutionary processes, thus enabling conservation decisions about a given species. We characterised the genetic structure of the Spanish Moon Moth Graellsia isabellae as revealed by multilocus (9 microsatellites) genotyping of 290 males sampled at the the Pyrenean National Park of Ordesa and surrounding buffer zone (a total of 13 sites separated by up to 28 km). Despite the overall low genetic differentiation and similar levels of diversity among sites, we found (i) significant west-east population structure as indicated by Bayesian clustering methods, (ii) a cryptic barrier to gene flow around one particular sampling site (Ordesa), and (iii) directional gene flow towards a second locality (Bujaruelo). Pairwise FST values showed that mountains prevent direct gene flow between the western and easternmost sites. However, low migrations rates were detected between intermediate sites (southern side of mountains), resulting in a clinal genetic variation throughout the Park, confirmed by isolation by distance analysis and individual membership coefficients to the clusters. We have analysed the patchy distribution and logging/reforestation history of the larval host plant (Pinus sylvestris) as explanatory factor for the cryptic barrier. Lastly, the combination of spatial configuration of woodland patches plus predominant wind direction is put forward to account for the directional migration toward Bujaruelo, a locality in the buffer zone near the National Park. Our work highlights the importance that buffer areas have to preserve high genetic diversity of forest insects within a national park.

MUTUAL EXCLUSION BETWEEN CRYPTIC SPECIES GENERATES MOST BETA-DIVERSITY IN WEST MEDITERRANEAN BUTTERFLIES

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The cryptic fraction of biodiversity is composed by morphologically similar species that are or have been overlooked by scientists. Most research is now directed towards documenting such cases, and estimating what will be the impact of this hidden fraction on the monumental task of describing Earth's biodiversity. However, both cryptic and non-cryptic fractions are generally assumed otherwise similar, and whether they may display different properties is a question that has not been specifically addressed. We use the butterfly fauna of the West Mediterranean to compare biogeographical patterns of the two fractions. We demonstrate that the cryptic fraction of diversity is overwhelmingly composed by groups of species that are not sympatric on mainland and do not co-occur on any island, and we specifically test for environmental and biogeographic hypotheses as the causes of such chequered distribution patterns. By analyzing the phylogeography and ecological preferences of two cryptic species pairs we show that neither dispersal capacity, nor climatic factors seem to explain the observed distributions, and that the existence of species interactions resulting in mutual exclusion is the most likely hypothesis. Finally, we demonstrate that mutual exclusion between cryptic species on islands accounts for most beta-diversity turnover in such areas. Thus, we show that the inclusion of cryptic diversity, frequently excluded in large-scale surveys and studies because of intrinsic identification difficulties, is of capital importance. The conclusion that the cryptic fraction of biodiversity displays unique and relevant gualitative features opens the door to study further potential particularities, as well as to assess their implications in multiple research fields.

POLYMORPHISM IN THE PAIN-1 VACUOLAR INVERTASE GENE IN SOLANUM SPECIES

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Invertases play an important role in carbohydrate metabolism. Moreover, invertases are involved in plant development and affect the formation of plant response to abiotic stress. In this work polymorphism in vacuolar invertase gene Pain_1 was characterized. Overall, 35 sequences of 28 species of the Solanum genus were analyzed. They included 9 tomato species and 11 potato species (subgenus Potatoe), other samples represented subgenera Leptostemonum, Minon, Solanum and Brevantherum.
The length of analyzed Pain-I exons III-V fragments varied from 591 to 962 bp as a result of multiple indels in intron III, which varied from 150 bp in S. peruvianum and S. habrochaites to 519 bp in S. pseudocapsicum. In total 80 indels were identified. The biggest deletions (157 bp) characterized intron III of S. sisymbriifolium, S. mauritianum, S. pseudocapsicum и Datura stramonium (taken as outgroups). Totally 188 SNPs were found. 49 SNPs were localized in coding regions. 39 of them resulted in 7 synonymous and 32 nonsynonymous aminoacid substitutions in the protein. Several substitutions and indels were specific to individual taxa or taxon groups, including potato and tomato species. Nine SNPs and eight allelic variants (SNP sets) have earlier been identified in exons III-V of S. tuberosum Pain-1. We examined the corresponding Pain_1 region in tuberproducing potato species (Solanum sect. Petota) and observed 6 new SNPs and 7 allelic variants. Two new allelic variants were found in the tomato species. In total, 18 new allelic variants were revealed in Solanaceae species examined. Obtained genetic distances and phylogenetic trees supported the commonly accepted taxonomic classification of the species, indicating that the Pain-1 fragment is suitable for taxonomic identification and phylogenetic studies in Solanaceae. This work was supported by RFBR grant 11-04-00446.

COMPARATIVE PHYLOGEOGRAPHY AND GENETIC POPULATION STRUCTURE OF TWO BUTTERFLY SPECIES FROM THE IBERIAN PENINSULA

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The Iberian Peninsula is recognized as one of the most important refugia for temperate species during the cyclic climatic changes of the Plio-Pleistocene glacial periods. Mountainous regions within these refugia areas are also believed to retain high levels of endemism and genetic diversity due to the easily tracked suitable habitats by upward-downward movements. Here we examine population genetic structure, phylogeographic and demographic patterns of *Lycaena bleusei*, a butterfly species endemic to the Central Iberian Mountain System and contrast this to the patterns found for a congeneric widespread species, also occurring in the Iberian Peninsula, *Lycaena tityrus*. We use two molecular markers to understand if there is congruency in their evolutionary histories or, conversely, if different evolutionary drivers may have shaped these species differently. We find that the two species, which exhibit similar dispersal abilities and ecological requirements, show different demographic and genetic patterns. We hypothesize that these differences are due to the evolutionary forces acting on species that contracted to southern refugia during the Pleistocene and that, ultimately, drove to population differentiation and speciation. We also find evidence for hybridization events between these two species, with *L. bleusei* males coming in contact with *L. tityrus* females on a secondary contact zone described here for the first time.

D21SY30PS1116

CRYPTIC GENETIC DIFFERENTIATION OF GAMBUSIA PUNCTICULATA (TELEOSTEI: POECILIIDAE) POPULATIONS ACROSS CUBA

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Diversification of freshwater fishes on islands has been considered unlikely because the traits that enable successful colonization, broad salinity tolerance and potential for oceanic dispersal, may also constrain post-colonization genetic differentiation. However some freshwater fish exhibit genetic differentiation and geographic structure on islands, whereas others do not. It is unclear what conditions give rise to contrasting patterns of differentiation because relatively few comparative reconstructions of population history have been carried out for insular freshwater fishes. Cuba is one of the main biodiversity hotspots of the Caribbean. 44 percent of the freshwater fish species are endemic, but their origin is poorly known. The family Poeciliidae is particularly relevant to study the relative contribution of colonization and local diversification because it includes more than 93% endemic species, some of them are tolerant to broad salinity levels and thus thought to be able to disperse through oceanic barriers. Local diversification is also expected, but it should be relatively low in an island with a relatively complex topology but potential connection between regions. We sampled 300 specimens of Gambusia puncticulata covering their geographic distribution in Cuba. We sequenced mitochondrial (Cytb, 12S) and nuclear (β-actin, S7) genes in order to reconstruct their phylogeny and their relationships with other non-Cuban species. 12 microsatellite markers were also analyzed. An important cryptic genetic diversity was observed which is concordant with previous results. In addition, it shows a high geographic organization in accordance with allopatric diversification on this island.

However, the close phylogenetic relationships between non-Cuban species with some Cuban lineages suggest multiple and independent events of colonization. We discuss the likelihood of these different scenarios in relation with the recent geological history of Cuba and Caribbean region.

MICROSCALE PHYLOGEOGRAPHICAL STUDY OF AGLAOCTENUS LAGOTIS (ARANEAE, LYCOSIDAE), A NEOTROPICAL WEB SPIDER

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The Atlantic forest extends along the Brazilian coast, characterized by a high species diversity and endemism. Around the city of Campinas (São Paulo) this forest covers two neighbor geomorphological units, the Atlantic Plateau and the Peripheral Depression, that, besides altitude, present different soil composition and microclimate conditions. Our aim was to investigate if populations of the two units might have different evolutionary histories. Aglaoctenus lagotis is a web builder spider, found in the Atlantic Forest as well as in the Brazilian savanna and Pantanal. We sampled 9 populations distributed over the two geomorphological units. A 521 bp region of the mitochondrial gene COI was sequenced for 96 individuals from the 9 populations and eight microsatellite loci were analyzed for five populations. The haplotype network revealed two groups separated by 13 mutational steps, one comprising the samples from Atlantic Plateau and the other the samples from Peripheral Depression. The microsatellite data corroborated the existence of the same two groups. The estimated molecular variability was similar for all populations. A bayesian phylogenetic tree was build and the estimation of age nodes used a strict molecular clock with mutation rate of 2,3%. All nodes dated in the Pleistocene: the divergence between the two groups was around 0,788 My; the radiation of populations from Peripheral Depression was later (~ 0,203 My) than the populations from Atlantic Plateau (~0,288 My), but the confidence intervals of these estimates overlap. The divergence of the groups may have occurred during a severe forest retraction but other hypotheses, such as different routes for the colonization of each unit cannot be discarded. Our data corroborate other findings of strong effects of the Pleistocene climate changes on the diversity of Atlantic Forest species.

CRYPTIC CRYPTO'S: USING A TARGETED DNA CAPTURE APPROACH FOR PHYLOGENOMIC ANALYSIS

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Elucidating the evolutionary relationships between lineages, is often a first requirement for studies focusing on micro- and macroevolutionary change. Where molecular phylogenetics has traditionally been hampered by the ability to generate large-scale orthologous sequence data, the recent developments in targeted capture of genomic regions in combination with next generation sequencing, now offer tremendous opportunity to generate such data and will aid in discerning phylogenetic associations at different levels of divergence. Here, we present the phylogenetic relationships between all currently described Australian *Cryptoblepharus spp.*, a genus of small skinks that inhabit a wide range of ecotypes but where species delimitation based on morphology is often confusing. Furthermore, initial mtDNA and allozyme analysis indicate that convergent evolution of phenotypes in similar environments seems to be reoccurring, regardless of phylogenetic association between lineages. To test this phylogenetic hypothesis, we used an exon capture approach to extract specific exonic regions and subsequently sequenced those on the Illumina platform, thereby creating a multi locus sequence dataset suitable for phylogenetic reconstruction. Our study demonstrates that exon capture followed by NGS of targeted regions, is a powerful approach for phylogenetic purposes and thus provides a reliable estimation of evolutionary history.

D21SY30PS1173

SPECIATION PATTERNS IN SKY ISLANDS – A CASE STUDY ON LEPTOPELIS FROGS FROM THE EASTERN ARC MOUNTAINS, TANZANIA

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The "sky island" system of the Eastern Arc Mountains is an ideal study area as it possesses high species diversity and has species with apparently widespread distributions across isolated mountains. The sky Island system therefore is an excellent area for studying speciation processes. We studied an amphibian model, *Leptopelis* tree frogs, which are restricted to montane forest habitats. We investigate the causal factors that might drive speciation processes and evolution of various phenotypes in the *Leptopelis* tree frogs. This is achieved by using a combination of phylogeographical approaches, phenotypic data, and fine scale spatial tools (GIS distribution modelling). We will present preliminary data on our findings.

RELAXED FUNCTIONAL CONSTRAINTS ON A TRIPLICATE ALPHA-GLOBIN GENE IN THE BANK VOLE

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Gene duplication plays an important role in the origin of evolutionary novelties, but the mechanisms responsible for the retention and subsequent divergence of newly created duplicates are not fully understood. The alpha-globin genes provide an example of gene family with different numbers of gene duplicates among rodents. While Rattus and Peromyscus each have three adult alpha-globin genes (HBA-T1, HBA-T2 and HBA-T3), Mus has only two copies. In Peromyscus, HBA-T3 appears transcriptionally inactive, despite the presence of an intact open reading frame and conserved splice junctions. We have cloned the alpha-globin genes from another cricetid rodent, the bank vole Clethrionomys glareolus, by RACE-PCR and assessed their relative expression by RNA-Seq. We show that the bank vole possesses three transcriptionally active alpha-globin genes with different expression levels. The HBA-T1 and HBA-T2 genes show 5-fold expression difference and are nearly identical in coding sequence. Although phylogenetically grouped with HBA-T1 and HBA-T2, HBA-T3 is distinguished by multiple amino-acid substitutions and is expressed 5- to 20-fold lower than HBA-T1 and HBA-T2. Alpha-globin polypetides corresponding to all three genes were detected by electrophoresis, suggesting that the bank vole HBA-T3 is a functional gene. Interestingly, we found two independent HBA-T3 alleles with a premature termination codon in exon 2 (PTC2) and exon 3 (PTC3), respectively. RT-PCR found evidence of transcription for the PTC3 allele, but not for the PTC2 allele. RNA-Seq reads uniquely aligning to the PTC2 allele suggest that its translation is reduced or prevented by nonsense-mediated mRNA decay (NMD). Due to the absence of a downstream exonexon junction, the PTC3 allele does not elicit NMD and is predicted to yield a truncated protein. The segregation of non-functional alleles in the bank vole suggests relaxed purifying selection and/or recent origin of HBA-T3 in this species.

D21SY30PS1192

RESOLVING SPECIES TREES AND REWRITING TAXONOMY: USING NEXT-GEN SEQUENCING DATA TO UNRAVEL A RECENT RADIATION OF A PASSERINE BIRD ON AN ISLAND SYSTEM

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Resolving species trees for island populations is challenging due to high complexity of demographic factors. We have analysed RAD tag SNPs from African Blue Tit (*Cyanistes teneriffae*) populations on the Canary Islands and from the mainland Blue Tit (*Cyanistes caeruleus*), applying only tools that do not rely on concatenation of loci. Our data provide resolution where earlier reports have been unsuccessful, and strongly indicate that the population on La Palma (*C. t. palmensis*) represent a recent colonization from mainland Europe while the other Canary Islands were colonized from Morocco, which seems to be the oldest population in our system. Sperm collected from these birds was used to test the hypothesis that divergence in sperm length mirrors genetic divergence, which held true against both SNPs and the mitochondrial COI marker. Our sperm data supports the phylogenetic division between *C. caeruleus* and *C. teneriffae*, and also *C. t. palmensis* as sister to mainland Europe. We conclude that recent advances in analytical tools make short-read sequencing technology a powerful option for phylogenetic analyses. Lastly we suggest that the taxonomy of *C. t. palmensis* be revised.

RELIABILITY OF ESTIMATING PHYLOGENIES USING MULTIDIMENSIONAL QUANTITATIVE CHARACTERS

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Quantitative multidimensional traits, such as shape, have been used in phylogeography and phylogenetics in recent years. However, there is no consensus about the reliability of the phylogenies built using these traits. Empirical studies have yielded different results in this regard, but no study has systematically studied it. Here, we simulate the evolution of four taxa under a Brownian motion model in a multidimensional space with varying dimensionality and different sets of branch lengths. In addition, two more sets of simulations with different degrees of integration among variables are tested using modified Brownian motion models. The percentage of correct phylogenetic reconstructions is used as a measure of phylogenetic accuracy. The results suggest that with low dimensionality, the phylogenetic accuracy is poor and problems of long-branch attraction appear. When few variables are used to describe the taxa, the reliability is poor and convergence appears even when two far-related groups of taxa are used, if one taxon within each group has evolved much. The accuracy is better when many variables are used. However, a high number of variables does not prevent from convergence when the variables used have the same pattern of dependency. Integration among variables has a strong negative effect. Reducing its effect implies knowing the pattern of interdependence among variables during the evolutionary process, which is difficult or impossible and requires large sample sizes. There are just very few favourable and restricted situations in which quantitative multidimensional data can be used to build phylogenies with accurate results. The effect of integration, really widespread, makes this technique not to be advisable.

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PROBLEMS WITH DNA-BASED SPECIES DELIMITATION WITH A BAYESIAN STATISTICAL APPROACH BECAUSE OF UPSTREAM ERRORS INVOLVING INDIVIDUAL ASSIGNMENTS TO PUTATIVE SPECIES

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Genetic-based delimitation of species typically involves a multistep process in which DNA data is analyzed with a sequence of different programs. Although the performance of the programs associated with each step have been evaluated separately, no analysis has considered how errors in the upstream assignment of individuals to putative species impacts the accuracy of delimited species in downstream analyses, such as those associated with the coalescent-based Bayesian statistical program bpp. Here we show that because of differences in the minimal data requirements for accurate performance in each of the different steps involved in the delimitation process, the reliability of inferences about species delimited from genetic data is compromised. Our results provide important insights into the practice of species delimitation by highlighting how uncertainty in the guide tree, as well as individual-species association (both derived from upstream analyses to bpp) can lead to under and overestimation of biodiversity, even though bpp itself may perform very well.

EXPOSING ADAPTIVE DIFFERENTIATION IN ANCIENT SCOTTISH PINEWOODS (*PINUS SYLVESTRIS* L.) VIA THE CONTROLLED APPLICATION OF WATER-STRESS

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The Scottish Highlands represent a small, but climatically diverse geographical area, and native pinewoods are found on sites with marked variation in altitude, seasonal precipitation, and soil drainage. We wish to determine whether native pinewood fragments are locally adapted to their environments, as the discovery of adaptive variation will benefit forest management and conservation policy, particularly in the midst of ongoing climate change. Presently, we are evaluating possible adaptive differentiation between native populations in response to soil waterlogging by means of a glasshouse based randomised block design progeny-provenance trial. Via the controlled application of stress, it may be possible to identify genetic differences between populations that would be otherwise elusive. The trial consists of 432 four year-old saplings originating from nine remnant populations located across an environmental gradient in Scotland. Half of the saplings have been waterlogged (continuously submerged to 1 cm below soil surface); half grown under normal watering. To acquire data at the population level, we have employed a fast physiological phenotyping technique to characterise stress responses to waterlogging in terms of photochemical capacity (via chlorophyll fluorescence). Additionally, by means of carbon isotope analysis, provenances have been screened for genetically-determined differences in water regime, both prior to and following the stress treatment.
The completed data set provides evidence that the Scotland's native pinewood remnants are genetically differentiated from one another, and furthermore that adaptive differences between populations in the glasshouse can be attributed to the prevailing conditions in their home provenances.

THE ROOT OF ALL WEEVILS: TOWARDS A TRIBAL-LEVEL MOLECULAR PHYLOGENY OF THE CURCULIONIDAE (INSECTA: COLEOPTERA)

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The true weevils are the most species-rich family-level taxon in the living world, numbering around 50,000 described species found across the globe in all terrestrial and subaquatic habitats where vegetation can grow. However, no fine-scale higher-level molecular phylogeny exists for this family as a whole, many of whose constituent subfamilies and tribes remain contentious because of widespread homoplasy in traditional morphological characters. Lack of a robust phylogeny has led to significant disagreements in weevil classification and taxonomy. This study aims to reconstruct the phylogeny of the Curculionidae at the level of the 292 currently recognized tribes and subtribes using molecular sequence data from multiple sources. The phylogeny is being achieved through a two-step protocol to maximise both taxon-sampling and sequence data (by utilising multiple mitochondrial and nuclear markers). Sequences from a long (c. 10Kb) portion of the mitochondrial genome were obtained using Long-Range PCR and next generation sequencing techniques from well-preserved specimens. These data were used to build a fundamental phylogenetic 'scaffold' comprising the main lineages, including most subfamilies. A further set of tribal representatives were sequenced for several nuclear and mitochondrial genes (18S, Arginine kinase, 28S, Histone 3, 16S, cox1) for finer resolution of the weevil tree. We also developed techniques for sequencing short phylogenetically informative shortamplicons from dried museum specimens, which typically contain fragmented DNA. The resulting phylogenetic hypotheses will help in uunderstanding the evolutionary history of morphological characters such as elytro-tergal stridulation and the rostral canal and receptacle, and the co-evolution and diversification of weevil lineages with specific angiosperm lineages. In addition, formal recognition of monophyletic lineages should lead to increased stability in weevil classification.

PHYLOGEOGRAPHIC ANALYSIS OF SPECIES OF THE FRESHWATER PSEUDOCRABS OF THE FAMILY AEGLIDAE OF SOUTHERN SOUTH AMERICA

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Aeglidae family is the only anomuran that live in freshwater environments of southern South America. The taxon is composed near 70 species and is not taxonomically resolved. The high endemism and diverse habitat colonized make that the Aeglidae species very particular taxa to study evolution. We analyzed mtDNA, COI-5P sequences (iBol Project) of seven species (Aegla scamosa, A. affinis, A. neuquensis, A. riolimayana, A. singularis, A. platensis, and A. uruguayana) that present inconsistencies in their morphological taxonomic characters. A Maximum Likelihood (ML) tree was performed by the software PHYLIP (v.3.69) and the consensus tree (by majority rule extended) was obtained Four species appear as monophyletic. However, A. neuquensis, A. singularis, and A. platensis show inconsistencies also observed in morphological studies. Most of the relationships among populations of each species were mainly correlated to their geographic distribution, and were not easily explained. Some populations were in doubt because their morphology and distribution could be considered new species or populations in process of speciation. La Blanca Lagoon specimens (A. affinis), a site isolated from the other hydrological habitats, appears as an out-group of the other A. affinis populations. Specimens of A. neuquensis have intermediate characters with A. riolimayana, and come out as an outgroup of sympatric specimens of Epulaquen Lagoons. Specimens of A. singularis from Uruguay River were clustered with specimens of A. platensis rather than populations of the same species but separated by a geographic barrier. A. uruguayana cluster, previously hypothesized as an ancestral population, come into view as the most recently. The biogeopgraphic patterns were similar as we had observed by geometric morphometrics analysis. These results shows Aeglids as an excellent model for study evolution due to the family particularities and the geo-climatic events occurs in the South America.

THE CARAPACE STRUCTURE OF FRESHWATER PSEUDOCRABS OF THE AEGLIDAE FAMILY. AN ADAPTATION OR EXAPTATION CASE?

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The Aeglidae family had its origin in freshwater environments 74 million years ago approximately. From that, the species in the family has suffered continuous environmental changes. In the present work we present a hypothesis about de carapace structure and their role in the evolution of the family. The structure selected was the carapace due to the multiple interpretations of its interaction in decapods. In addition to the fact that its shape is a distinctive feature of the taxa, species presents lines and grooves whose origin and function are still under debate. This pattern of lines and grooves divides the carapace into discrete units (modules), and appear as an autapomorphy of a genus of uncertain origin and functions. Some authors discuss the function for such lines and grooves as representative of evolutionary ancestral limits, whereas others put forward that some carapace grooves in decapods are secondary structures. Specimens of different species of the Aeglidae Family were photographed and 31 landmarks were selected representing the carapace whose was divided into 5 modules according the lines and grooves. The modules were compared among them intraspecifically and interspecifically and different degrees of variation were observed. Intraspecific analysis reveals that the modules, corresponding to anterior region of the pseudocrabs, present greater variation than those of the posterior region of the carapace. While interspecifically, the pattern of shape variation are the same and statistical significance were observed among the same modules. Then, the evidence analyzed could be interpreted as modules like by product of adaptation in changing environments or exaptation to other functional structures, as different pressures of others appendices or internal organs.

SPATIAL AND TEMPORAL SCALES OF EVOLUTION OF A PIGMENTATION PHENOTYPE IN CICHLID FISHES

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Cichlid fishes are an excellent model system for speciation studies for their multiple radiations, in particular in East African lakes. In Lake Tanganyika, the most successful lineage of cichlids is the Lamprologini. One group of lamprologines, the *Neolamprologus brichardi/pulcher* complex, or Princess of Burundi, is found throughout the rocky shores of Lake Tanganyika. Different species occupy multiple discontinuous patches of coastal habitat and are characterized by differences in facial stripe pigmentation. Such pigmentation is relevant for intraspecific communication and previous results based on mtDNA data from 3 species suggest parallel evolution of facial stripe phenotypes. We assess the spatial and temporal scales of differentiation and speciation in this group of cichlids by analyzing mitochondrial and nuclear patterns of variation in 11 species and several hundreds of individuals originating at multiple localities from distinct patches of their distribution. We contrast these genetic and geographic data with facial stripe phenotypes to understand the evolution of this pigmentation trait in *Neolamprologus*.

SEMI-PERMEABLE SPECIES BOUNDARIES AND TIME-DEPENDENT REPRODUCTIVE ISOLATION IN IBERIAN BARBUS AND LUCIOBARBUS (TELEOSTEI, CYPRINIDAE)

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The study of species boundaries provides valuable information on how biodiversity is generated and maintained, but is complicated by several factors that influence the association of genetic and morphological variation with specific taxa. As taxa diverge, ancestral polymorphisms get sorted and independently evolved traits become correlated, allowing us to discriminate among different forms. Nevertheless, shared traits can often be observed among otherwise distinct taxa, reflecting recent common ancestry and incomplete sorting of characters, introgressive hybridization, or a combination of both. The two processes are difficult to separate, but the patterns they generate can potentially be discriminated using multiple markers and geographical information. The impact of these processes on the evolution of species boundaries is most easily studied in recently and rapidly radiating groups that show some degree of range overlap. Taxa that present these ideal characteristics are the speciose genera Barbus and Luciobarbus. Here we examine different classes of characters (external meristic traits, mtDNA and nuclear DNA) and their patterns of covariation in endemic Iberian barbels sampled from sympatric and allopatric populations. We show that species boundaries are permeable to gene flow, as sympatric populations of different species show an increased number of shared alleles compared to their allopatric counterparts. Furthermore, the degree of reproductive isolation is heterogeneous throughout the genome, since distinct nuclear markers show different levels of introgression. Finally, isolation seems to spread along the genome with the establishment of new mutations, as more distantly related species are more reproductively isolated than closely related ones.

UNRAVELLING LANDSCAPE FACTORS SHAPING SPATIAL GENETIC STRUCTURES OF SPECIES INVOLVED IN A SPECIFIC PLANT-INSECT MUTUALISM

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Nursery pollination is one of the most fascinating examples of plant-pollinator interactions, in which the pollinator also acts as a seed parasite. In Europe, only few species evolved towards this strategy, with the notable example of the European globeflower Trollius europaeus and its exclusive fly pollinators in the genus Chiastocheta (Diptera: Anthomyiidae). In this interaction the plant is exclusively pollinated by flies, whereas in turn larvae eat a fraction of the developed seeds. Because of the relatively simple nature of this interaction (one plant and a small set of pollinators), this system can serve as a model for studying how topography influences genetic diversity of mutualistic species. This question is of major importance in evolutionary biology as plants and insects generally show different population parameters associated with contrasting life-history traits, and as a consequence, different spatio-temporal dynamics, challenging maintenance of specificity in a coevolutionary framework. The novelty of the study also lies in the combination of data sampled from two trophic levels at different spatial scales and in four replicates, using specimens collected from four mountain massifs, i.e. the Sudetes, Tatra (Poland), Alps, and Jura (Switzerland). As the genus Chiastocheta encompasses seven species, our approach also allows comparing the spatial genetic structures of different pollinator species, which, despite being closely related, sympatric over large areas, and ecologically similar, differ in natural history traits such as oviposition patterns and resource exploitation strategies. Finally this study evaluates the level of congruence in spatial genetic structures between the insects and the plant, allowing examination of the extent to which plant gene flow is driven by pollinators' dispersal patterns rather than by seed dispersal.

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TIME SCALES OF ANURAN SPECIATION INFERRED FROM HYBRID ZONES OF PALEARCTIC GREEN TOADS (*BUFO VIRIDIS* SUBGROUP) IN A PHYLOGEOGRAPHIC FRAMEWORK

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A central assumption in evolutionary biology is that reproductive isolation accumulates with increasing genetic distance and thus divergence time. In anuran amphibians, experimental breeding might still produce viable offspring after >20 My divergence. However, the timing of speciation in closely related anuran lineages is poorly studied. Green toads with differently old forms in secondary contact offer an ideal system. In a phylogeographic framework (>300 locs., >700 mtDNAs), we molecularly examine divergence times, and interactions in three natural hybrid zones (>150 populations; 12 msats, mtDNA). Under Plio-Pleistocene (2.7 Mya) divergence (Sicily: B. siculus, B. balearicus), we demonstrated advanced reproductive isolation with bidirectional mtDNA (40 km) but scarce nuclear introgression. Pairwise FST maxima, but genetic diversity (He) minima at the contact suggested absence of current gene flow. In northeastern Italy, we studied interactions between younger lineages of early Pleistocene (1.9 Mya) divergence (B. balearicus, B. viridis) that pose a striking contrast. We found significantly greater nuclear (40 km) and wider asymmetric, differential mtDNA introgression (100 km), with a slight He increase at the centre, and pairwise FST changing smoothly across the hybrid zone. FST maxima occur in most apart populations, mirror isolation by distance and a significant effect of dispersal barriers (Po, Adige). After middle Pleistocene divergence (1.2 Mya), in Greece (B. variabilis, B. viridis; work in progr.), a uniform mtDNA lineage of Asia Minor variabilis has colonized the viridis

refugium on the Balkan Peninsula. MtDNA and nuDNA-hybridization stretch >200 km. All of this reflects profound differences in hybridization between lineages from the same radiation, which are mostly distinguished by evolutionary age. Our study is in line with research on other anuran species but has the advantage to compare differently diverged lineages from the same radiation.

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SPATIAL GENETICS OF INFECTIOUS DISEASE; IDENTIFYING MULTIPLE UNKNOWN SOURCES

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One approach to landscape genetics is to describe the terrain as being comprised of a patchwork of genetically distinct populations. We present an alternative perspective, which treats each individual as having dispersed across the landscape from a source; that source being one of a number of alternatives. We argue that this methodology is much more effective in the analysis of biological invasions, the spread of infectious diseases, and related problems. We build a Dirichlet process model incorporating both spatial and genetic information, and demonstrate some of the useful outputs that can be obtained from this model structure. As well as inferring the likely location of the unknown sources, the model allows the actual number of sources to be directly estimated. Computational innovations provide the efficiency required to allow for biologically realistic (non-normal) dispersal, as well as a variety of mutation models.

D21SY30PS1306

SPECIES LIMITS AND PHYLOGEOGRAPHY OF THE LEPTIDEA CRYPTIC SPECIES COMPLEX (LEPIDOPTERA: PIERIDAE)

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Although European butterflies are among the best studied insects on the planet, cryptic species are still being discovered. The common and widespread Wood White butterfly L. sinapis was among the first cases shown to represent two different species (L. sinapis and L. reali). However, very recently, it has been discovered that yet another cryptic species, L. juvernica, has been hidden within L. reali. The presence of such surprising levels of cryptic diversity in an already intensively studied system, makes the Leptidea triplet a unique case among European butterflies and prompted us to study the species limits and phylogeography of the cryptic species complex. Through extensive mating experiments, we found that all three species are reproductively isolated due to female mate choice of exclusively conspecific males. The three species are closely related but differentiated genetically. They have homogeneous intraspecific genetic structures suggesting that their current distributions are the result of a postglacial colonization from a single glacial refugium for each species. The only differentiated lineage are the Irish L. juvernica, that may have survived in southern Ireland during the last glacial maximum. Recent findings indicate that *L. sinapis* is the species with the highest known chromosome number variability (2n=56-106), excluding cases of polyploidy. Interestingly, the variation in chromosome numbers follows a longitudinally-oriented cline. This context offers the rare possibility to test the concept of clinal species. Our preliminary results based on mating between different chromosomal races of *L. sinapis* suggest that pronounced differences in karyotypes associated with multiple chromosomal rearrangements are not strongly detrimental in this species.

D21SY30PS1313

PRELUDE TO THE STORY OF DOMESTIC GOOSE IN EUROPE

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The development of next generation sequencing based techniques has increased the amount of data available for population genetic studies. NGS techniques have also become popular in the studies of domestication due to genome-wide information they can provide on the species of interest. Chicken has been the most popular object of study on avian domestication due to its worldwide distribution. We wanted to focus on a more unfamiliar yet important domestic bird species, that is, the European domestic goose. We used a genotyping by sequencing (GBS) approach to detect single nucleotide polymorphisms in domestic geese and its wild ancestor, the greylag (Anser anser). We consider the impacts of inbreeding, drift, bottlenecks and artificial selection in promoting differentiation between the wild and domestic birds. We discuss the potential of GBS to investigate other aspects of the story of domestication of the goose such as where and when it happened.

CRYPTIC DIVERGENCE AND EVOLUTIONARY CONVERGENCE IN THE DIVERSIFICATION OF THE SONGBIRD GENUS JUNCO (AVES: EMBERIZIDAE)

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The rate and magnitude of phenotypic evolution can vary greatly among traits and taxa, and cases of convergent evolution and mutation-order divergence can further confound lineage history and the inference of speciation processes. We use molecular genetic tools and patterns of phenotypic variation to reconstruct the evolutionary history of the songbird genus Junco in order to understand the factors, mechanisms and timing of their diversification across North America. Analysis of mtDNA sequence and AFLPs revealed that plumage diversity in the dark-eyed junco, which has puzzled ornithologists for over a century, evolved recently as the yellow-eyed junco colonized the North American continent from southern Mexico following the Last Glacial Maximum. A new analysis of all known junco forms using a multilocus dataset reveals that junco populations at the tip of Baja California, Guadalupe Island in the Pacific, and the highlands of Guatemala, represent divergent lineages that have been isolated for hundreds of thousands of years, yet have differentiated relatively little in most traits. A phylogeny of the group reveals that the yellow-eyed and dark-eyed juncos are paraphyletic taxa. The Guadalupe junco is an old evolutionary lineage whose similarity to mainland dark-eyed juncos in plumage and eye color is due to convergence. Some phenotypic traits (eye and several plumage color traits) are not phylogenetically informative in juncos, whereas bill color or song characters are more consistent with neutral genetic markers. Drift or mutation-order divergence in long-term geographic isolation likely explains the differentiation of Guatemala and Baja juncos. In contrast, a role for sexual selection must be invoked to explain the rapid diversification of continental dark-eyed junco taxa. New genomic markers are being developed in order to clarify phylogenetic relationships among incipient lineages, identify "divergence islands", and detect specific regions under selection.

D21SY30PS1347

THE EFFECT OF RECOMBINATION ON POPULATION DIFFERENTIATION

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Evolution can be viewed as the movement of populations through a space of genotypes. This space can be modeled as a network connecting genotypes accessible through mutation. Here I investigate the evolutionary dynamics in the simplest version of the genotype network model, where all genotypes in the network have the same fitness (the neutral network model), combined with a finite island model of population subdivision. I find that the structure of the genotype network determines whether there is a positive or negative correlation between recombination rate and population differentiation at equilibrium.

DELINEATION OF THE SAND FLY (DIPTERA: PSYCHODIDAE) SPECIES WITHIN THE 'MAJOR GROUP' OF TURKEY

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Members of the subgenus Larroussius are the only proven vectors in the Mediterranean region of Leishmania infantum, the causative agent of visceral and cutaneous leishmaniasis in humans and canines. Sand fly species of this subgenus are grouped into four species complexes, namely Phlebotomus perniciosus, Phlebotomus perfiliewi, Phlebotomus ariasi and Phlebotomus major, which are considerably sympatric in their range of distribution from South West Asia to Northern Africa and Southern Europe. Among these complexes, the taxonomic status and distribution of the morphologically similar members of the Phlebotomus major complex in Turkey are unclear. To examine the utility of traditional morphological characters and molecular markers, sand flies were sampled from 90 localities in eleven different provinces covering a wide geographical range throughout Turkey. The morphometric variability was analyzed using multivariate analyses of twelve characters, while mitochondrial cytochrome b (Cyt b) and nuclear Elongation Factor 1α (EF 1- α) genes were used for molecular discrimination. Three distinct monophyletic lineages were identified based on the phylogenetic analysis of the combined data set of mitochondrial and nuclear gene regions, which were also supported by parsimony haplotype network analysis and AMOVA of Cyt b. The first lineage is restricted to south eastern Turkey and represents the species Phlebotomus syriacus, the second is present mostly in the westernmost and the easternmost localities and represents P. neglectus, and the third member of this complex is distributed across the mid-northern and mid-southern regions. None of the studied morphological characters were found to be sufficient to discriminate between these three members of the P. major s.l. complex; however their presence sympatrically in several localities supports their status as species rather than inter-population variability.

CRYPTIC DIVERSITY IN *PTERONOTUS PARNELLII* (MORMOOPIDAE) FROM THE GUIANA SHIELD: EVIDENCES FROM MOLECULAR, MORPHOMETRIC AND BIOACOUSTIC TOOLS

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Mormoopidae is a neotropical family of bats with two genera, Mormoops and Pteronotus, ranging from south Texas to Brazil, and west of the Andes to Peru. The last systematic revision of this group recognized six species within Pteronotus: parnellii, macleavii, quadridens, personatus, davyi and gymnonotus. In P. parnellii, the most abundant and widely distributed species, numerous subspecies have been recognized along its distribution, but for the geographic range of the Guiana shield just the subspecies P. p. rubiginosus is actually described. Interest in the evolution and genetic variation of P. parnellii has increased recently and molecular studies have evidenced a cryptic diversity within this taxon. We investigated the diversity in P. parnellii based on acoustic (n=200), molecular (n=80) and cranial (n=80) tools in Guyana, Suriname, French Guiana and Brazil. The acoustic analysis showed two distinct groups in the same area: Group 1 emitted with a Frequency of Maximal Energy (FME) around 53 kHz, and Group 2 had their FME around 59 kHz. The skull measurements evidenced that Group 1 animals are larger than Group 2 ones, with several cranial variables allowing a discrimination of the two sympatric taxa. In the phylogenetic results, two distinct lineages with a genetic divergence higher than 5% were found. These lineages clearly correspond to the groups found by previous analyses, being composed by the same individuals. The groups are sympatric in part of their geographic distribution, co-occurring in many localities. Our results indicate that Group 1 corresponds to P. p. rubiginosus by including all specimens sampled from Mato Grosso, Brazil, the type-locality of this subspecies. It occurs from Central Brazil to the Guianas. Group 2 is found in the Brazilian states of Pará and Amapá, and in the Guianas. Based on multiple evidences, we believe that Groups 1 and 2 represent two distinct species, which we denote *P. rubiginosus* and *Pteronotus sp nov*, respectively.

UNEXPECTED GENETIC POPULATION STRUCTURE IN THE KEA (NESTOR NOTABILIS)

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The Kea (Nestor notabilis) is an endemic alpine parrot of the South Island of New Zealand. After 150 years of persecution, it was fully protected in 1986, but the species is still declining over its whole range. Here I present data from 17 microsatellite and one mitochondrial markers for Kea sampled at nine different locations along the length of the South Island. Kea are known to have considerable dispersal capabilities and are sometimes sighted far from their alpine habitat. It is therefore expected that such a potential for gene flow would make distant populations less genetically differentiated. However, we found a significant isolation by distance (IBD) pattern. Moreover three distinct genetic clusters were identified using microsatellite data while mitochondrial data identified two main groups of haplotypes. Our data suggest that kea population might have been reduced to at least one refugium during the last glaciations and subsequently recolonized their range. We discuss possible causes for this unexpected population structure, including social behavior and call differences as a barrier to dispersal, the "beech-gap" hypothesis and potential implications for species management.

EVOLUTION OF MUTT PROTEIN AND THE EMERGENCE OF THE AEROBIC WORD

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The MutT protein family has been considered as an important protection against DNA damage and play an important role in protecting living form against oxidative stress in the aerobic word. Here, I have examined the evolution of this family under a new hypothesis of its function: the protection of the GTP binding protein from guanine oxidized nucleotide, 8-oxo-GTP. A phylogenetic tree was constructed based on the protein sequences of mutT members from different kingdoms. The phylogenetic analysis suggested that mutT gene evolved very early in the life history when the first organisms began to face de novo generated aerobic conditions. The evolutionary tree of MutT proteins also suggests, under the light of new findings, that this family was essential for the life evolution in the news aerobic times.

D21SY30PS1400

SPECIATION BEYOND EXPECTATION: THE EVOLUTION OF LAPAROCERUS (CURCULIONIDAE) IN THE CANARY ISLANDS

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On a given island or archipelago, the proliferation of live has progressed much farther within some lineages than others. For example, among the 88 native genera of weevil within the Canary Islands, 128 species, more than one third of all described species, belong to a single genus, Laparocerus. The important question is not only why diversification occurs on islands, but why do some lineages diversify so extensively on islands, while others do not? As a first approach to understand why Laparocerus has diversified dramatically within the Canary Islands, we have sampled 176 individuals within the Laparocerus tessellatus species complex from across its distribution. Using mtDNA and nuclear ITS2 sequence data, we first ask to what extent geography structures genetic variation within species. To evaluate this we (i) characterise the geographic ranges of mitochondrial and nuclear alleles, and (ii) assess the extent to which alleles of recent common ancestry share similar geographic ranges, and (iii) test for geographic signal in patterns of genotypic relatedness. We then ask to what extent patterns of genetic relatedness among species reveal evolutionary process, in particular genetic admixture. To evaluate this we (i) identify alleles of recent common ancestry that are shared between species, and (ii) use a geographic framework for speciation to evaluate hypotheses of incomplete lineage sorting against gene flow. Both gene regions indicate an origin for the complex on Gran Canaria, and both reveal geographic structuring of genetic variation within islands. An unexpected result is that mtDNA data reveals each of the single species on LaPalma and ElHierro to be the product of more than one colonization event from more than one source island. In both cases nuclear data reveals these multiple colonisations to have been followed by genomic admixture. These results raise interesting possibilities for genomic admixture and evolutionary success within the genus Laparocerus*

D21SY30PS1447

COMPARING PATTERNS OF GENETIC DIFFERENTIATION BETWEEN ISLAND AND MAINLAND POPULATIONS OF TWO MOUNTAIN NEWTS

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Island populations are expected to show reduced genetic diversity, population structure, and coalescence times with respect to mainland populations. We compared the phylogeographic and population genetic structure of two mountain newts, one endemic to the Corsica island (*Euproctus montanus*), and one endemic to the Apennine chain (northern Italy; *Mesotriton alpestris apuanus*). The two mountain ranges are close to one another (less than 200km far), have largely overlapping latitudinal ranges, and experienced similar climatic conditions throughout the Pleistocene. Contrary to the expectations based on its insularity, *E. montanus* showed far deeper phylogeographic structure and higher genetic diversity than the continental species *M. alpestris*. Indeed, we found five phylogroups of Pliocene origin among *E. montanus* populations, whereas three phylogroups of Pleistocene origin were found in *M. alpestris*. Differences in species response to past climatic oscillations. However, these data, together with the growing literature on intra-island genetic variation, suggest caution with the use of simplifying assumptions on the evolutionary history and demographic structure of island populations, particularly as far as conservation planning is concerned.

D21SY30PS1452

PHYLOGEOGRAPHIC ANALYSIS OF THE MEXICAN FRESHWATER FISH COMPLEX CATOSTOMUS NEBULIFERUS-PLEBEIUS

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The complex Catostomus nebuliferus-plebeius is endemic group of fish Northwestern Mexico (C. nebuliferus y C. plebeius), member of the Catostomidae family distributed in North America. The complex is distributed in basins of Sierra Madre Occidental: *Catostomus nebuliferus* in the Nazas river; while *C. plebeius* is distributed in the Mezquital, Piaxtla, Fuerte, Yaqui, Conchos and internal basins derived from the Guzmán lagoon. Therefore the origin and activity of this mountain system should be associated to the evolutionary history of the complex. The first plausible scenario is where this mountain serves as a center of diversification. The second scenario would be that the current distribution of complex is resulted of admixture between populations isolated in the lowlands to the west and east of the mountain during the Pleistocene interglacials states. We implemented phylogeographic, phylogenetic and population genetics analysis to describe the evolutionary history of complex. High genetics distances were found in preliminary results between all populations of the complex (3-5% with Cytochrome *b* mitochondrial gene) and high genetic diversity in each population, indicative of an ancient isolation. A process of isolation occurred ~5.7 millions years ago in the Pliocene, isolating the population of Nazas and Mezquital of the North basins, resulted of the geological events of the region; while in the same period (~4.5 millions years ago) were separated populations from Mezquital river and *C. nebuliferus* populations. The populations of *C. plebeius* of the north basins were isolated gradually during the last 4 millions years describing a South-North isolation pattern associated to a geological activity. Null migration rate was estimated between populations after isolation discarding of river piracy in the Pleistocene. Our results have taxonomic and conservation implications, because the complex currently consists of several independent evolutionary lineages.

EVOLUTION OF POLYPLOIDY AND FUNCTIONAL DIPLOIDIZATION IN STURGEON: MICROSATELLITE ANALYSIS IN TEN STURGEON SPECIES

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Sturgeons are 'living fossils' among fishes and one of the most endangered groups of animals. Multiple polyploidization events make this group unique subject for studying evolution of polyploidy in animals. As most sturgeon species have gone through significant functional diploidization, two scales of ploidy levels can be distinguished: the 'evolutionary scale', which indicates the maximum ploidy level achieved, and the 'recent scale', which indicates current functional ploidy level. In this study we used microsatellite analysis to determine ploidy level and degree of functional diploidization in 10 sturgeon species from Europe and Asia. Maximal number of alleles per individual of a given species was assessed at 20 microsatellite loci. Our data on the percentage of disomic loci in different species suggest that functional diploidization is ongoing process in sturgeon. We observed lower level of diploidization in tetraploid species from the Atlantic clade than in the species from the Pacific clade, which can be explained by more recent genome duplication in tetraploid species from the Atlantic clade. Conservation management suffers a lack of appropriate DNA markers suitable for identification and description of hybridization in sturgeon species of different ploidy level, especially in Danube Delta. Only three diploid - Huso huso, Acipenser stellatus, A. ruthenus and one tetraploid sturgeon species - A. queldenstaedtii are still reproducing in the Lower Danube River. They are threatened by hybridization among them, and with another non-native tetraploid species - A. baerii. As microsatellite markers are most suitable for hybrid description, we tried to detect microsatellite loci with disomic inheritance in polyploid species, A. gueldenstaedtii and A. baerii. While very low percentage of the markers showed disomic pattern in these two species, several of the markers appeared useful for A. stellatus, A. ruthenus and H. huso.

D21SY30PS1465

DIVERSITY, HOST ASSOCIATIONS AND PHYLOGEOGRAPHY OF HYPOMYCES (HYPOCREALES, ASCOMYCOTA) CAUSING THE COBWEB DISEASE OF WILD AND CULTIVATED MUSHROOMS

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Hypomyces represents the largest genus of ascomycetes parasitising on fruitbodies of fungi. Temperate species producing red pigments and having *Cladobotryum* anamorphs are common on species from five orders of Agaricomycetes. Isolates causing the cobweb disease of commercially cultivated mushrooms result in serious economic losses. We sequenced five gene regions in 90 wild strains and 30 strains from mushroom farms, isolated from Europe, North America, Africa, Asia, Australia and New Zealand. Multigene analyses reveal that five species include causal agents of the cobweb disease for which the name *H. rosellus* has widely been misused. The diversity in local species pool determines the species composition in mushroom farms of the same region. The majority of such isolates belong to *H*. odoratus, including a group of fungicide-resistant strains from Europe and North America sharing identical sequence data. Our results support the distinctness of five species but reveal the core species, H. rosellus to be paraphyletic, comprising six cryptic lineages. Hypomyces rosellus s. str. is characterised by wide dispersal and gene flow across Eurasia but does not occur in North America. Instead, the lineages from the West and the East Coast of USA appear distinct, having given rise to species inhabiting temperate Australasia. Separate lineages appear to be maintained by geographic isolation in these regions but by host specialisation in the species occurring sympatrically in Europe and Asia. Both specialist and generalist host use strategies have evolved in the group. The prevalence of clonal reproduction suggested for H. odoratus, could account for its success in mushroom farms facilitating the spread of fungicide resistance. tef1 and rpb2 can be recommended for species delimitation. Low variation, not exceeding 1% in the whole ingroup, argues against the use of ITS rDNA, the universal DNA barcode marker for fungi, for species recognition in this group.

Symposium

31. Development, Behaviour and Evolution

22, 23 and 24 August



Program

Thursday 22 August

Session(s): 9

Friday 23 August

Session(s): 10, 11, 12, 13

Saturday 24 August

Session(s): 14

Organisers: Élio Sucena and Christen Mirth

Invited speakers: Cristina Ledon-Rettig, Laura Corley Lavine, Wiliam

Jeffery and Emilie Snell-Rood

Description:

Evolution moulds developmental processes to create the diversity of morphologies, life history strategies and behaviours displayed across organisms. Recent developments in areas such as genomics and developmental biology promises to further broaden our perspectives of this topic. This symposium aims to bring together investigators working on various aspects of the development, behaviour and evolution of phenotypes to highlight exciting new avenues of research and stimulate the exchange of ideas.

D22SY31IT10:30R4

LEARNING-LIKE MECHANISMS ACROSS DEVELOPMENT: INSIGHTS FOR THE EVOLUTION OF LIFE HISTORIES AND COMPLEX TRAITS

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A long standing question in biology is understanding how complex and diverse traits can arise out of a relatively simply genome. Developmental selection, a process analogous to learning, has long been touted as a mechanism that can explain the emergence of complexity in the immune and nervous systems. Here, I argue that a broader appreciation of developmental selection across all traits is necessary for understanding phenotypic evolution. In particular, developmental selection increases the likelihood of adaptive plasticity and integrated complexity in traits, but comes with energetic and time costs relative to a specialist, resulting in major changes in life history strategies. In this talk I review evidence that developmental selection applies to a range of traits, from behavior to gene expression, and suggest that incorporating this mechanism into our evolutionary models will lead to a more complete understanding of innovation, diversification and complexity.

D23SY31IT15:45R1

DEVELOPMENTAL LINKS BETWEEN SEX AND NUTRITION; INTERACTIONS BETWEEN THE SEX DETERMINATION PATHWAY AND HORMONAL SIGNALING IN STAG BEETLES

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The evolution of sexual dimorphism in animals has long fascinated biologists. One of the important characteristics of sexually-selected traits is condition dependent expression. The mandibles of metallic stag beetles are sexually dimorphic and disproportionately larger in males with large body sizes compared to small body size males. The development of these sexually selected traits is likely to be regulated by nutrition-dependent mechanisms as well as the sex-determination pathway. Yet we know very little about how either of these mechanisms work, much less how they might interact with each other to generate sexual dimorphism. In this study, we experimentally offer evidence of a developmental link between nutrition and sex in the sexually dimorphic stag beetle, Cyclommatus metallifer, focusing on genes in the sex-determination pathway and its interaction with juvenile hormone (JH) signaling which acts as a mediator between nutrition and mandible growth.

D23SY31IT17:45R1

EVOLUTION OF A NOVEL ADAPTIVE BEHAVIOR MAY PROMOTE EYE REGRESSION IN ASTYANAX CAVEFISH

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The teleost Astyanax mexicanus is a species complex consisting of sighted surface-dwelling (surface fish) and blind cave-dwelling (cavefish) forms. Cavefish show constructive phenotypes, such as novel behaviors mediated by enhanced sensory systems, and regressive phenotypes, including the loss of eyes and pigmentation. Because surface fish and cavefish are interfertile genetic approaches can be used to study these phenotypes. Vibration attraction behavior (VAB) is a foraging behavior that detects aquatic vibrations emanating from living prey and requires a specialized class of sensory neuromasts (orbital neuromasts or ON). VAB is pervasive and strong in cavefish but is only rarely seen and weak in surface fish. Genetic analysis of F2 progeny obtained by crossing surface fish with cavefish showed that VAB is paternally inherited and controlled by several QTL that are congruent with QTL for ON number. Importantly, two of the major VAB and ON QTL cover the same genomic regions as some of the QTL responsible for cavefish eye degeneration. This QTL clustering, along with negative phenotypic correlations between VAB/ON and eye size in the F2 generation, suggest that the underlying constructive and regressive phenotypes are co-regulated, either by antagonistic effects of the same pleiotropic gene or by tightly linked genes that are subject to genetic "hitchhiking". Thus the evolution of eye loss in cavefish may be a consequence of strong positive selection for VAB and its sensory receptors, whose enhancement is adaptive in the dark cave environment.

D24SY31IT10:30R2

ALTERED DEVELOPMENTAL PROGRAMS UNDERLYING THE NOVEL GUT MORPHOLOGY OF A CANNIBALISTIC ANURAN LARVA

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The gut is a particularly fascinating organ from an ecological and evolutionary perspective; it dictates how well an organism digests and assimilates nutrients from its diet, and thus has a profound impact on fitness. Organisms vary widely in what they consume, and this dietary diversity is reflected by equally diverse variation in gut morphologies. For instance, anuran *Xenopus laevis* larvae maintain an elongate gut that is adaptive for their herbivorous diet (an ancestral condition). In contrast, larvae of Lepidobatrachus laevis are obligate carnivores (a derived condition) and possess a relatively short gut that is appropriate for their diet. Remarkably, little is known about how developmental programs have diverged to give rise to such variation in gut morphology. To reveal the developmental mechanisms underlying the evolution of the novel, short-gut morphology, we compared key features of gut morphogenesis between Xenopus and Lepidobatrachus. In Xenopus, all endodermal cells of the primitive gut tube become polarized, undergo radial intercalation and contribute to gut elongation as they become incorporated into the gut epithelium. In contrast, in Lepidobatrachus, many central endodermal cells do not undergo radial intercalation and therefore cannot contribute to elongation. Additionally, many cells in the developing epithelium of *Lepidobatrachus* undergo programmed cell death and are extruded, further limiting the endodermal contribution to elongation. This programmed cell death – which typically occurs during thyroid hormone-dependent metamorphosis in *Xenopus* – is correlated with the expression of thyroid hormone signaling components in Lepidobatrachus, suggesting that precocious thyroid hormone signaling in the developing intestine is, in part, responsible for their novel gut topology.

D22SY31RT11:18R4

SEX-SPECIFIC EFFECTS OF PATHOGENIC BACTERIAL INFECTION ON FITNESS COMPONENTS, REPRODUCTIVE BEHAVIOUR AND MATE CHOICE IN DROSOPHILA MELANOGASTER

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We investigated the sex specific effect of Serratia marcescens infection through septic injury on two components of fitness, survivorship and adult competitive reproductive fitness in male and female Drosophila melanogaster. Under competitive conditions, infected females showed significantly lower reproductive output compared to control females. However, male reproductive fitness was not affected by infection. We did not find any cost of injury and immune deployment in our study indicating that the fitness cost resulted only from sustained infection or virulence. However, the bacteria was found to be pathogenic, growing equally well in males and females and causing about 80% mortality over an eleven day period with no significant difference in mortality between sexes. Thus, such sex specific effects of pathogens on individual components of fitness indicate that the total fitness cost experienced by each sex might be very different, even within a given regime, thereby affecting the evolution of antibacterial immunity. Given such sex-specific effects of bacterial infection on reproductive fitness, we further, investigated the sex-specific effect of bacterial infection on components of reproductive behaviour. We did not find any change in mating latency or copulation duration with male or female infection status. When given a simultaneous choice between infected and sham infected females, males preferentially mated with sham infected females. This result suggests that male *D. melanogaster* may adaptively bias their mating effort in response to the infection status of females.

D22SY31RT11:42R4

EVOLUTIONARY PATHWAYS TO CONVERGENCE IN PLUMAGE PATTERN PHENOTYPES

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Convergence in animal phenotypes is a central theme in evolution. Bird plumage patterns are an emerging model system due to the hierarchical nature of feather growth and spectacular within-feather pigmentation. Although birds are visually diverse, at the level of individual feathers there are just three major types of regularly repeating patterns: scales, bars and spots. Previous studies of avian pattern phenotypes have predominantly been adaptationist, focusing on camouflage and/or communication. In contrast, developmental constraint in plumage pattern convergence is relatively unexplored. A biologically realistic model of within-feather pattern formation (Prum & Williamson 2002) relies on a reaction-diffusion based mechanism. According to this model, scales require the least stringent regulation, bars require a higher rate of activation decay than scales, and spots require simultaneous spatial and temporal differentiation. Thus, development may constraint scales to evolve first, followed by bars and finally spots. Here, we use Bayesian phylogenetic modeling to reconstruct the evolutionary pathways to plumage pattern convergence, in the two avian clades with the most spectacular plumage patterns – waterfowl (Anseriformes – 118 species), and gamebirds (Galliformes – 170 species). Plumage pattern evolution was allowed to evolve independently in six feather tracts over the body, in each group separately. We found that for each tract, in both groups, scales predominantly evolve first, followed by barred and spotted plumage. Interestingly, we found that spotted plumage is more likely to evolve from scaled than barred plumage in gamebirds, but not in waterfowl. Together our results demonstrate directionality in plumage pattern evolution, providing support for developmental constraint that may be mediated by reaction-diffusion dynamics. These results underscore the importance of considering selection in the context of development in studies of phenotypic evolution.

D23SY31RT10:30R9

CORRELATED EVOLUTION OF LEARNING RATE STUDIED BY ARTIFICIAL SELECTION IN A PARASITIC WASP

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Learning is considered a form of behavioral plasticity through which an organism can adjust its behavior to changes in the environment. Because of both the apparent costs involved in learning and the fact that the rate of learning is highly context-dependent (depending on reliability of certain cues, the value of the reward, the expected lifespan of the organism) we expect and observe many speciesspecific learning rates. Even closely related species can demonstrate quite different learning rates and differences in consolidation rates of memory for similar experiences. These rates are therefore most likely adapted to the needs of the organism in its natural environment. Because many aspects of learning and memory formation share rather conservative regulatory pathways, rates of learning related to different types of behavior can be correlated (e.g. overall high learning rates for different stimuli). We explored this concept in the parasitic wasp Nasonia vitripennis by creating an artificial selection line favoring a high learning rate for associating a color with finding a host (after a single associative training trial). This selection regime selects for a higher rate of forming a type of middle-term memory. We then explored whether the established 'high-learning' lines also exhibit a correlated higher learning rate for a novelty stimulus like odor and whether formation of other memory types (short term or long term memory) was also affected. Also, changes in a selection of life history traits like fecundity and developmental rate were considered, as well as genetic differences. Here I will present the first results of this study.

D23SY31RT10:54R9

METABOLIC RATE AND BODY SIZE LINKED WITH PERCEPTION OF TEMPORAL INFORMATION

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Body size and metabolic rate both fundamentally constrain how species interact with their environment and are hence an important determinant of the niche they occupy. While many mechanisms linking how both intra- and inter-specific interactions are constrained by body mass and metabolic rate have been explored, their effects on the fundamentally important resolution at which temporal information is perceived have been largely overlooked. The visual system acts as a gateway to the dynamic environment and the relative resolution at which organisms are able to acquire and process visual information is likely to restrict their ability to interact with events around them. As both smaller size and higher metabolism are known to facilitate rapid behavioral responses, we hypothesize that these traits will favor perception of temporal change over finer time scales. Using the critical flicker fusion frequency, the lowest frequency of flashing at which a flickering light source is perceived as constant, as a measure of the maximum rate of temporal information processing in the visual system, we present a phylogenetic comparative analysis of a wide range of vertebrates supporting this hypothesis. Our analyses show that species that have small body masses, high metabolic rates and live in bright environments have the highest temporal perceptual abilities. Our results have implications for the evolution of signaling systems and predator-prey interactions, and, combined with the strong influence that both body mass and metabolism have on a species' ecological niche, suggest that time-perception may constitute an important and over-looked dimension of niche differentiation.

D23SY31RT11:18R9

NEUROGENETIC NETWORKS AS SUBSTRATES FOR NATURAL VARIATION IN OLFACTORY BEHAVIOUR IN DROSOPHILA MELANOGASTER

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Appropriate responses to chemical signals are essential for survival and reproduction, and genetic variation that gives rise to phenotypic variation in olfactory behaviour provides a substrate for natural selection. Drosophila melanogaster provides an ideal system to study the genetic basis for natural variation in olfactory behaviour because its olfactory system is well characterized and genetically identical individuals can be reared in controlled environments. The Drosophila melanogaster Genetic Reference Panel (DGRP), a collection of inbred wild-derived lines with fully sequenced genomes enables GWA studies to be performed in a scenario where all variants are known, candidate genes can be functionally tested allowing empirical assessment of the FDR, and outbred populations derived from DGRP lines can be constructed for the analysis of epistasis. GWA analysis together with variance GWA (vGWA) analysis in the DGRP revealed single nucleotide polymorphisms (SNPs) associated with variation in olfactory behaviour. Subsequent extreme QTL mapping using advanced intercross line (AIL) populations derived from lines with opposite extreme behavioural responses revealed additional SNPs. Combined analyses of GWA, vGWA and extreme QTL mapping uncovered a cellular network associated with variation in olfactory behaviour, centered on genes involved with cellular signaling and neural development. Candidate genes and network connectivity were validated through mutational analysis. We show that different elements of the genetic architecture underlying natural variation in olfactory behaviour are revealed in GWA studies and extreme QTL mapping contingent on allele frequencies and context-dependent effects, but they converge on similar cellular processes. Such genetic architecture appears to be a general feature of quantitative traits, is likely to be universal, and has profound implications for the interpretation of human GWA studies and the evolution of complex traits.

D23SY31RT11:42R9

TESTING EVOLUTIONARY MODELS FOR THE MAINTENANCE OF SAME-SEX SEXUAL BEHAVIOUR

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The evolutionary maintenance of same-sex sexual behaviour (SSB) is an enduring mystery because of its apparent negative impact on reproductive fitness. Two influential genetic models of SSB were developed several years ago by Sergey Gavrilets and William Rice: SSB might be maintained by sexually antagonistic selection for fitness-increasing traits in the opposite sex, and/or it might result from overdominance if alleles enhancing its expression are maintained by a heterozygote advantage. These models make distinct predictions about the genetic architecture of SSB and its correlations with male and female fitness. We present results of an experiment to empirically test these predictions using the Drosophila Genome Reference Panel. We screened 50 highly-inbred, genome-sequenced D. melanogaster lines for male SSB and performed a series of crosses between high and low lines designed to discriminate the two hypotheses. There was substantial additive genetic variation for SSB, strong effects of the social environment, and evidence for G x E. We will discuss genetic variance components contributing to this behaviour, candidate loci found to be strongly associated with its expression, context-dependence of the influence of those loci, and effects of SSB on fitness of males and females. Taken together, the results enable us to statistically compare support for either hypothesis - sexual antagonism vs. overdominance - and provide an intriguing contrast with Drosophila knockout studies that have found a wide assortment of genes that might contribute to variation in male SSB.

D23SY31RT14:00R9

A TALE OF TWO ENDS: AXIS POLARITY IN THE SCUTTLE FLY MEGASELIA ABDITA

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In dipterans (flies, midges and mosquitoes), segment determination occurs early in embryogenesis through a hierarchy of genes activated by maternal factors. These include the anterior determinant bicoid (bcd), found only in Cyclorrhapha or "higher" flies, and also caudal (cad), hunchback (hb), and nanos (nos). Loss of any one of these genes in Drosophila results in the loss of segments, and in some cases duplication of anterior or posterior structures at the opposite pole of the egg. However, in none of these cases is global polarity lost. In contrast, removal of both maternal *bcd* and *hb* results in symmetrical embryos (mirror-abdomen or bicaudal phenotypes) in which global anterior-posterior polarity is lost. Similar bicaudal phenotypes can be generated in two other fly species, the hover fly Episyrphus balteatus and the scuttle fly Megaselia abdita, by knocking down only maternal bcd. In *Episyrphus*, this has been explained by a lack of maternal *hb*. However, in *Megaselia* this factor is present so another explanation must be found. We have generated gene knock-downs for each of the maternal factors in *Megaselia* and analysed the resulting gene expression patterns. We identify loss of Kr activation by maternal hb as the likely cause of the loss of polarity. Furthermore, we identify differences in *cad* and gap gene expression that account for the more anterior plane of symmetry observed in Megaselia bicaudal phenotypes. These differences suggest that a number of regulatory changes in the gap gene network have occurred since the divergence of these dipteran lineages. We are testing these inferred changes by performing more knock-down experiments, combined with datadriven modeling of the gap gene system in this species. Finally, we revisit the question of *Kr* activation in *Drosophila* and suggest an evolutionary scenario for the development of anterior-posterior polarity in dipterans.

D23SY31RT14:24R9

MAJOR CHANGES IN THE CORE DEVELOPMENTAL PATHWAYS OF NEMATODES: ROMANOMERMIS CULICIVORAX REVEALS THE DERIVED STATUS OF THE CAENORHABDITIS ELEGANS MODEL

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Despite its status as a model organism, the development of *Caenorhabditis elegans* is not necessarily archetypical for nematodes. The phylum Nematoda is divided into the Chromadorea (indcluding C. *elegans*) and the Enoplea. Compared to *C. elegans*, enoplean nematodes have very different patterns of cell division and determination in development, while at the same time the adult vermiform phenotype is extremely similar across the phylum. Embryogenesis of the enoplean *Romanomermis culicivorax* has been studied in great detail, but the genetic circuitry underpinning its development is unknown. We created a draft genome of *R*. *culicivorax* and compared its developmental gene content with that of two nematodes, C. elegans and Trichinella spiralis (another enoplean), and a representative arthropod Tribolium castaneum. This genome evidence shows that R. culicivorax retains components of the conserved metazoan developmental toolkit lost in C. elegans. T. spiralis has independently lost even more of the toolkit than has *C. elegans*. However, the *C. elegans* toolkit is not simply depauperate, as many genes essential for embryogenesis in C. elegans are unique to this lineage, or have only extremely divergent homologues in R. culicivorax and T. spiralis. These data imply fundamental differences in the genetic programmes for early cell specification, inductive interactions, vulva formation and sex determination. Thus nematodes, despite their apparent morphological conservatism, have evolved major differences in the molecular logic of their development. R. culicivorax serves as a tractable, contrasting model to *C. elegans* for understanding divergent genomic and thus regulatory backgrounds generating a conserved phenotype. The draft genome will promote use of R. culicivorax as a research model.

D23SY31RT14:48R9

GENERATING MORPHOLOGICAL VARIATION FROM BASIC DEVELOPMENTAL PROCESS: THE EXAMPLE OF THE DROSOPHILA WING

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Understanding the genotype to phenotype map (G-P map) is a central problem in evolutionary biology. The G-P map determines the phenotypes that can be produced, and structures the possible evolutionary responses to selection. The complexity of both the genotype and the phenotype may suggest that the map concept is of little practical value in predicting either the range of possible phenotypic variation, or the way that traits evolve. We argue that this difficulty may be overcome by focusing on the developmental level. While the molecular basis of morphological variation is indeed complex, there are only a few morphogenetic developmental mechanisms (e.g., cell proliferation, movement, orientation of cell division, etc) that generate morphology. Understanding on the one hand how the spatial and temporal distribution of transcription and growth factors affect these mechanisms, and on the other hand, how variation in these mechanisms affects morphology provides a practical approach of the G-P map. To illustrate this approach, we built a cell-based computational model of Drosophila wing development that links cell signalling and biomechanics and wing morphology. The model represents cells as 2D polygons. Cell movement, division, biomechanical properties, and cell-cell interactions are implemented together with realistic cell signalling (wingless, dpp, vestigial) and intracellular gene networks. The model generates the 2D morphology of the wing, veins, cell position, shape and polarity. We will present the range of in silico morphological variation obtained through the exploration of the parameter space of the model. This variation will be confronted with the morphological variation obtained in the lab by quantitatively manipulating the level of expression of key developmental genes.

D23SY31RT15:12R9

GENETIC EVIDENCE FOR MOSAIC BRAIN EVOLUTION IN MAMMALS

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The mammalian brain consists of distinct parts that serve different functions. It has been argued that evolution of the mammalian brain is constrained by developmental programs (concerted evolution) and that different brain parts are not free to respond individually to selection and evolve independent of other parts as predicted by a mosaic scenario of brain evolution. We test these hypotheses using a quantitative genetic approach involving over 10,000 mice and identify independent loci for size variation in seven key parts of the brain. We further demonstrate that brain parts show low or no phenotypic correlation, as is predicted by a mosaic scenario. We also demonstrate that variation in brain size is independently regulated from body size. The allometric relations seen at higher phylogenetic levels are thus unlikely to be the product of strong developmental constraints.

D23SY31RT16:33R1

QUANTITATIVE TRAIT LOCI FOR BEHAVIOR, GROWTH AND BODY SIZE IN THE NINE-SPINED STICKLEBACK PUNGITIUS PUNGITIUS L.

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Genetic architecture of ecologically important traits, such as behavior and body size, are as yet relatively little studied in wild animals. Using F2-intercross (n = 283 offspring) between behaviorally and growth strategically divergent nine-spined stickleback (Pungitius pungitius) populations, we explored the genetic underpinnings of growth and behavioral traits describing different aspects of activity and boldness with the aid of quantitative trait locus (QTL) analyses based on 226 microsatellite markers. The behaviors were analyzed separately (viz. feeding activity, risk-taking, exploration) and also in combination to map "behavioral types". Significant (experiment wide) QTLs were detected for both trait groups. In addition, suggestive (chromosome wide) QTLs were also detected. The results also showed that loci affecting size and growth traits were located in some cases in the same chromosomal region as loci affecting behavior of the nine-spined stickleback. The results found in these studies lay the foundations for fine mapping these traits and provide a starting point for identification genes responsible for the size and behavior differences between marine and pond nine-spined sticklebacks.

D23SY31RT16:57R1

REVISTING GAVIN DE BEER'S CLASSIC EXAMPLE OF HETEROCHRONY: THE DEVELOPMENTAL BASIS OF JAW GROWTH IN HALFBEAKS AND NEEDLEFISH

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Heterochronic shifts in growth can generate novel, adaptive phenotypes that facilitate access to previously unoccupied niches. As they can arise from simple developmental switches, encoded by few mutations, novelties that arise through heterochronic shifts, are ideal systems for investigating the molecular basis of evolutionary change. Fishes from the suborder Belonioidei achieved a vast array of craniofacial morphologies that appear to have arisen through a series of heterochronic shifts. In some species (needlefishes) both the upper and lower jaws are highly elongated in others only the lower jaw (half-beaks). We examined the molecular basis of accelerated heterochronic growth in the lower jaw, relative to the upper jaw in the half beak Dermogenys pusilla, and compared it to a their sister group of the suborder the medaka, where this trait is ancestral, i.e., both jaws are equally long. Previous investigations have identified Calmodulin as a likely molecular determinant of accelerated jaw growth in D. pusilla, as its developmental expression reflects the relative growth of the jaws, while expression remains equal between the upper and lower jaws in medaka. Here we describe new transcriptome-wide analyses identify further candidates for accelerated jaw growth in the halfbeak, which also shed light on the structural composition of the tissues that comprise this morphological novelty. Furthermore, comparative gene expression analyses in medaka provide insights into the ancestral condition, enabling further assessments of mutational changes that may underlie a major heterochronic shift that is shared amongst the Belonoidei. As this shift appears to have been critical enabling the Belonoidei to occupy a greater number of trophic niches than the medaka, we postulate that this heterochronic shift explain the considerably greater evolutionary success of the Belonoidei compared to the medaka lineage.

D23SY31RT18:33R1

THE MOLECULAR BASIS OF A NOVEL PIGMENT TRAIT IN CICHLID FISHES

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How novel traits emerge and are modified is one of the many unresolved questions in evolutionary biology. The spectacularly diverse adaptive radiations of cichlid fishes in the East Africa Great Lakes provide an ideal system to study the molecular basis of evolutionary novelties in the context of adaptation and explosive speciation. One characteristic innovation of the most species-rich lineage of cichlids, the haplochromines, is a set of brightly pigmented spots on male anal fins, known as "eggspots". Egg-spots are a diverse trait (number, colour and shape), and this trait plays a key role in the territorial and breeding behaviour of about 1,500 species of cichlids. Here we report the identification of several egg-spot candidate genes by quantitative next generation sequencing of RNA from egg-spot tissue in the haplochromine cichlid Astatotilapia burtoni. We confirmed these results in other haplochromine species through quantitative gene expression analysis (qPCR), and narrowed down our study to one gene – an androgen receptor (AR) cofactor. A comparative genomic analysis between haplochromines and egg-spot-less non-haplochromine species reveals that the coding region of this gene cannot explain the origin and diversity of this trait. However, the upstream regulatory region of the AR cofactor differs between these groups: haplochromines bear a unique transposable element insertion in the proximity of the transcription initiation site of the AR cofactor. We designed constructs containing this AR-cofactor transposable element region upstream of a GFP (green fluorescent protein) coding sequence and tested them in zebrafish. GFP expression was detected in iridophores (pigment cells) and in the fin rays. We thus propose that this transposable element insertion might have changed the expression pattern of AR-cofactor, thereby initiating the morphogenesis of a key evolutionary innovation in one of the most species-rich lineages of vertebrates.

D23SY31RT18:57R1

VISUAL ADAPTATION OF THREESPINE STICKLEBACK (GASTEROSTEUS ACULEATUS) TO DIVERGENT SPECTRAL REGIMES

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Vision is a sensory modality of fundamental importance for many animal species, aiding in foraging, the detection of predators and colour based mate choice. Consequently, adaptation to divergent spectral environments is often essential and can influence reproductive isolation. This study collected opsin gene expression data from 11 populations of threespine stickleback (Gasterosteus aculeatus) in order to model the spectral sensitivity of stickleback inhabiting divergent spectral environments. We found that the spectral sensitivity of freshwater stickleback populations was significantly shifted toward longer wavelengths relative to their marine ancestors. The observed divergence in spectral sensitivity was repeated for multiple independently derived populations inhabiting similar spectral habitats, suggesting parallel evolution of the visual system and the action of natural selection. This shift in sensitivity has important implications for prey detection and mate choice. Correspondingly, divergence in the visual system and requires further attention. Additionally, the observed shifts in visual sensitivity after freshwater colonization from marine environments could be extremely widespread.

D24SY31RT11:18R2

PREDATORY CANNIBALISM IN DROSOPHILA MELANOGASTER LARVAE

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Predatory cannibalism involves hunting live conspecifics of size similar to (or larger than) oneself. Although this is risky and is thought to require specialized behavioural and morphological adaptations similar to those in predatory animals, instances of such predatory cannibalism have been reported in numerous non-carnivorous species. It remains unclear if such predatory cannibalism has adaptive significance, while research into its genetic and sensory basis has been hindered by the lack of a suitable model system. Here we report predatory cannibalistic behaviour in *Drosophila melanogaster* larvae and address its evolutionary significance. We found that groups of younger larvae regularly attack and consume larger healthy conspecific larvae. Aggregations of cannibalistic larvae are mediated by strong attraction to chemical cues from the attacked victim. The nutrition obtained through cannibalism is significant: we show that *Drosophila* can complete their egg-to-adult development on an exclusively cannibalistic diet, and that this diet induces phenotypic plasticity of the mouth hooks. Finally, during 118 generations of experimental evolution, replicated populations maintained under larval malnutrition evolved enhanced propensity towards cannibalism, in addition to changes in their larval-foraging path length (sitter-like behavior) and competitive ability. These results indicate that predation on conspecifics in *Drosophila* involves specific adaptations, has a survival value, and can rapidly evolve in response to nutritional conditions. Thus, even in non-carnivorous animals natural selection can promote cannibalistic behaviour. This discovery additionally changes the perspective on many aspects of behaviour, ecology and evolution of Drosophila and simultaneously provides an extensive model for investigating the neuronal, ecological and evolutionary aspects of cannibalistic behaviour.

D24SY31RT11:42R2

COMPETITION DRIVEN ECOLOGICAL SPECIATION IN LAKE TANGANYIKA CICHLIDS

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Disruptive selection operating on different habitat use can lead to ecological speciation. However the selective forces are often unclear. In Lake Tanganyika the cichlid fish *Telmatochromis temporalis* has two morphs: a small-bodied "shell" breeding morph, and a large-bodied "rock" breeding morph. We first show that body size differences are heritable in laboratory-reared first generation offspring. Using mtDNA and AFLP data we then show populations are philopatic, and that these morphs have evolved repeatedly. We next tested for divergence on habitat preference using substrate choice trials on wild-caught fish. We found individual habitat preferences in the absence of other fish were tightly linked to body size with larger individuals using rock, and smaller individuals using shells, irrespective of collection habitat. However, medium-sized "shell" males also used rock when given a free choice, but the presence of large competitor "rock" males caused them to shift towards the less preferred shell habitat, and to pair assortatively. Thus, the results support competition as a crucial agent driving the dichotomous habitat but outcompeted on rocks, while larger fish are selected against on shell habitats by predators.

POSTERS

D23SY31PS0048

PERSONALITY AND LIFE HISTORY TRADE-OFFS IN WANDERING ALBATROSS

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Trade-offs are central to life history theory and frequently reported between current and future reproduction. As a result individuals display polymorphic strategies across their lifetime but the proximate causes driving individual variation remain largely unknown. Theoretical models propose life history trade-offs as both a causal factor and mechanism maintaining animal personalities and there is increasing empirical evidence linking personality to individual life history traits. In this study we extend these analyses, to ask whether personality may predict an individual's lifetime reproductive strategy, or "pace of life". It is predicted that "fast" living individuals breed early and invest heavily in each attempt, whereas "slow" individuals delay reproduction, invest more strategically over time, but experience reduced or delayed senescence. Here, quantifying the change in life history traits across the lifetime of individuals, we test whether personality predicts an individual's investment and pace of life. Using personality data from over 1100 wandering albatross and demographic data collected across 47 years, we demonstrate that variation in early life investment is predominantly driven by sex specific strategies, but senescence in later life is strongly dependent on both personality and sex. We report sexually antagonistic effects, with bold females and shy males suffering early and rapid senescence in reproductive success. Effects are mainly apparent in hatching success and we examine whether differences in foraging behaviour may drive the observed patterns. We discuss the implications of our results for individual senescence and consider how sexually antagonistic selection and differences in lifetime fitness may affect selection on personality in this population.

D23SY31PS0057

HOW FAR DO WASPS FLY AND HOW DO THEY RETURN HOME?

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Ropalidia marginata is a primitively eusocial, independent founding, polistine wasp widely distributed in peninsular India. Although we have a fair amount of information about its nesting biology and social organization, we know little about its abilities and mechanisms of orientation, navigation and homing. In an attempt to understand how far the wasps fly and how they manage to return to the nest, we removed and transported foragers from 4 nests, without giving them a view of the surroundings, and released them in 4 cardinal directions, in gradually increasing distances from the nests. Only foragers were used for the experiment because they were already observed to fly out and successfully return to their nests. All wasps (43/43 wasps and a total of 198/198 releases) successfully returned to their nests when they were released within an area of about one sq. km around their nests. We also released 125 wasps beyond an area of one sq. km around their nests but within four sq. km around their nests. Here the wasps returned sometimes but not always: of the 243 releases, wasps returned only in 140 cases. No wasps (0/64 wasps) returned to their nests when they were released beyond four sq. km around their nests. In another experiment, when all wasps, including foragers and non-foragers were released at a point 100 meters away from their nests, only some (38/64 wasps) successfully returned to their nests, even when they were given a view of their surroundings during transportation. In this experiment, the probability that a wasp returned to her nest was positively correlated with the proportion of time that she had spent away from her nest prior to the experiment. These results are consistent with the hypothesis that familiarity with the surroundings is both necessary and sufficient for the wasps to perform successful homing.

D23SY31PS0082

POTENTIAL PITFALLS OF CROSS-FOSTERING TO TEST PARENT-OFFSPRING CONFLICT AND COADAPTATION

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The often complex interplay between offspring begging for food and food-providing parents is a key feature of the family life of many animal species. Theory predicts that this interplay may ultimately lead to a coadaptation between parental provisioning and offspring begging, which will be stabilized by fitness costs to parents and offspring. There is now mounting evidence which supports such predictions for a number of species, including the canary (Serinus canaria), our model species. These studies typically used cross-fostering designs in order to control for the potential effects of the provisioning of parents on the begging of their biological offspring and vice versa. However, it is still unknown to what extent cross-fostering per se changes the expression of these behaviours and which costs may be associated with cross-fostering. Thus, we investigated whether (or not) offspring begging is affected by the cross-fostering and estimated the potential associated costs for the offspring in terms of growth and survival. Offspring begging and parental provisioning levels were tested, both on day 3 (= before crossfostering a subgroup of broods) and day 5 after hatching (= 2 days after cross-fostering a subgroup of broods). The begging intensity was lower at day 5, but this change was not different between control and cross-fostered broods. Neither did we find fine scale adjustments of the offspring to the parental qualities of the foster parents (estimated from their provisioning behaviour). In addition, cross-fostering had -on average- no negative effects on offspring growth and survival. Thus, these results suggest that cross-fostering does not entail costs to the offspring, despite the fact that chicks do not respond to the change in parental qualities.

D23SY31PS0096

USES AND ABUSES OF THE TERM CONFLICT IN DEVELOPMENTAL BIOLOGY AND EVOLUTIONARY MEDICINE

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The term conflict has been used widely in evolutionary biology to describe outcomes of competition over resources between genetically different individuals; e.g. rivals, mates, relatives. The parentoffspring conflict theory, introduced by Trivers in 1974 and furthered by Haig in the 1990s, describes metaphorical conflict between maternal and offspring genes (and paternal and maternal genes in an offspring). A developing offspring shares 50% of its genes with its mother but 100% of its genes with itself, thus the conflict theory predicts that a fetus demanding more resources than is in the mother's interest to give will increase its own (and its father's) fitness at the mothers expense. Conflict theory has been applied to explain aspects of developmental physiology including evolution of the placenta, genomic imprinting, and maternal constraint. However, many original predictions made by the conflict theory are not supported by recent empirical and theoretical evidence. The conflict theory also has been applied in ways to suggest that the (metaphorical) conflict between maternal and fetal genes can be unproblematically extended to the level of the organism. Yet, particularly in humans, in many of these cases cooperation, coadaptation, and conflict resolution appear to be better explanations than 'conflict" itself. While application of conflict theory has led to insights into the developmental relationship and application of evolutionary theory to the human condition is essential, a more integrated developmental paradigm that also includes resource availability, mother's age and reproductive history, mono/polytocous birth, multiple paternity, gestation length, and fetal phenotype response to the environment, is vital in defining the maternal fetal relationship. Those in different fields should use caution, understanding, and perspective when applying concepts such as genetic conflict and distinguish between conflicts at different levels of selection in future work.

D23SY31PS0121

WING PIGMENTATION AND THE EVOLUTION OF WING SHAPE

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Wing shape is affected by different selection pressures. For instance, when wings have pigmentation, they can be involved in colour signalling for sexual selection. Therefore, pigmented wings may show morphological variations not expected by aerodynamic predictions alone. We present results from a series of comparative studies where we explored the role of wing pigmentation in the evolution and diversification of a group of pigmented-winged damselflies. Pigmentation is a sexually selected trait related to individual male fitness in these damselflies. At the intraspecific level, we found that wing shape is correlated with the expression of wing pigmentation, presumably related to male ability to maintain a territory. At the interspecific level, we found that the presence and extension of pigmentations were related to wing shape variation in hindwings, but not in forewings. Wings showed larger regions where the pigmentation was located. Moreover, we also found a faster evolution of hindwings in comparison to forewings, which fits with the major role of hindwings in signalling during courtship flights. Interestingly, we found that the differences in wing shape are not related to different levels of flight agility between species. Our results suggest a correlated evolution of hindwing shape and wing pigmentation in order to improve colour signalling to potential mates. In our system, evolution of wing shape seems to be partially decoupled, between fore- and hindwings. Finally, despite different gender-specific strategies and clear sexual dimorphism in wing shape, we also show evidence for correlated evolution of wing shape between the sexes.

D23SY31PS0131

SOCIAL CONFLICT IN ANT LARVAE: A CASE FOR CANNIBALISM

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In social organisms cooperation is widespread, however social groups also provide excellent opportunities for individuals to exploit the cooperative efforts of others, thus creating conflict. Ants provide some of the most intriguing examples of social conflict in nature, for example when colony members differ in their interests concerning male parentage. Such conflicts are often mediated by kinship, and the ability of individuals to assess kinship within colonies plays an important role in conflict resolution. However, whilst much is known about conflict in adult ants, the social role of larvae has been largely neglected. We investigated a novel social conflict in ants by measuring competition among larvae in the form of egg cannibalism. We found that first instar larvae of the ant F. aquilonia eat eggs and that levels of cannibalism differed depending on the origin of eggs. While 10% of larvae ate sibling eggs, 24% of larvae ate eggs from a foreign population. Discriminatory behaviour matched population-specific egg odours, providing first evidence that larvae detect and react to chemical cues in a similar way as adult ants. Cannibalistic larvae showed prolonged survival compared to non-cannibals, indicating that cannibalism bears benefits for ant larvae. We also found that males cannibalised significantly more often than females, which points to sex-dependent selection for selfish behaviour. These results suggest that larvae are not the powerless individuals they have been seen as so far. Instead, developing offspring and male larvae in particular may be important players in social conflict in ants, and social animals in general.

D23SY31PS0163

IT SMELLS LIKE BLUE

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In nature, animals are exposed to a wide variety of sensory cues, auditory, olfactory, visual or gustatory. These sensory cues may be used as information to learn to recognize dangers and resources in a changing environment. In order to associate a neutral stimulus to an aversive or appetitive one (associative learning), information may be collected by different sensory systems, which may vary in their reliability and may even be in conflict. How is processed information? Is there information transfer between sensory systems? How animals balance these different informations will have a strong impact on their decision making. Using *Drosophila melanogaster*, a well-known model for learning ability studies, we propose to understand how different sensory modalities (gustatory, visual and olfactory) can be used in an associative learning task and how they interact.

D23SY31PS0237

GENETICS OF SEX DETERMINATION AND SEXUAL TRAITS IN THE PARASITOID WASP GENUS ASOBARA

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Sex determination and sexual-reproduction traits are key issues in evolutionary biology. One way to improve our understanding of sex determination is to study the underlying mechanisms in closely related species and in a phylogenetic framework. In the Hymenopteran parasitoid wasp genus Asobara, characterized by a haplodiploid sex determination system, we first investigated if haplodiploidy in four different species is caused by complementary sex determination (CSD), whereby sex differentiation depends on the allelic state at the CSD locus (heterozygous individuals are females, hemi- or homozygous are males). Using multiple-generations inbreeding experiments, flow cytometry and simulation models, we showed that sex in this genus is neither determined by the allelic state at a single locus nor at multiple loci, but by an alternative mechanism. We then focused on the species A. japonica which, in addition to sexual lineages, comprises asexual lineages as a result of Wolbachia infection. It is expected that in asexuals, traits once involved in sexual reproduction become redundant and vestigialize. To gain insights into the genetics of sexual vestigialization, genes from an asexual lineage were introgressed into a sexual lineage, by taking advantage of males that are occasionally produced by asexual females. Among the offspring of F1 hybrids, approximately 50% of females lost their mating propensity, suggesting a simple genetic architecture underlying trait decay, consistent with a single recessive locus. Sex allocation among the females that did mate became more female-biased in F1 generation and slightly less female-biased in further introgressed generations, suggesting a complex genetic architecture with dominance effects. Using next-generation sequencing of different Asobara species, we are investigating the molecular basis of sex determination mechanisms and vestigialized sexual traits. I will present and discuss the data in an evolutionary context.

D23SY31PS0244

LIFE-HISTORY PATHWAYS ASSOCIATED WITH SEQUENTIAL REPRODUCTIVE TACTICS IN THE PEACOCK BLENNY (SALARIA PAVO)

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A polymorphism has been documented in males of the peacock blenny (Blenniidae: Teleostei) according to which males can reproduce either as parental males (bourgeois males that express secondary sex characters such as a head crest and a sex-pheromone producing anal gland) or as sneaker males (smaller and younger males that lack male secondary sex characters and mimic female courtship behaviour). In this study the hypothesis that the two male morphs correspond to different life-history pathways was tested by following marked individuals in the field (Ria Formosa, Portugal) during a period that covers their life span (6 years). Two main pathways were identified: 1. early hatched immature males become bourgeois males at age 0+/1 and reproduce as such in their first reproductive season and subsequent ones ("Bourgeois pathway"); 2. later hatched males reproduce as sneakers in their first reproductive season switching to the bourgeois tactic after the end of that season, at the start of age 1, and they reproduce as bourgeois males in the subsequent seasons ("Sneaker pathway").. These results confirm that the pattern of alternative tactics is flexible with fixed sequences, and supports the "birthdate effect" as a factor influencing tactic expression. The estimated reproductive success of the "bourgeois pathway" was lower than that of the "sneaker pathway". Therefore early hatching may constitute a constraint that we hypothesise to be maintained by an opposing selective force that favours early hatching of females.

D23SY31PS0258

NO SPACE FOR A LARGE BRAIN? TESTING CONSTRUCTIONAL CONSTRAINTS ON BRAIN SIZE EVOLUTION IN LAKE TANGANYIKA CICHLIDS

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Brain size is enormously variable among vertebrates and this variation is thought to be maintained through the balance of adaptations, trade-offs, and constraints. Although intuitive in theory, the impact on brain size evolution from constructional constraints exerted by head morphology has so far been largely overlooked. We used geometric morphometrics on the morphologically diverse Tanganyikan cichlids to investigate whether head morphology co-evolves with brain size. We examined the relationship between head shape and brain size while controlling for phylogenetic relationships and potentially confounding ecological factors. The analyses revealed significant associations between head shape and brain size. We found that species with curved, wide and high heads had large brains while narrow pointed head profiles were associated with small brains. This finding supports the hypothesis that constructional constraints can play a role in brain evolution. Our result, together with previous works on other groups of vertebrates, suggests that eco-morphological adaptation to specific feeding technique might cause constructional constraints on vertebrate brain evolution.

D23SY31PS0266

INVASIVE CRAYFISH HAVE A GREATER IMPACT ON AMPHIBIAN LARVAL SURVIVAL AND POND STRUCTURE THAN NATIVE PREDATORS

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The efficiency of induced antipredator responses critically depends upon accurate cue recognition and hence requires some joint predator-prey evolutionary history because otherwise preys may fail to recognize or respond efficiently against alien predators. Thus, amphibian larvae often fail to trigger antipredator responses against invasive predators. Few studies analyzed the ecological consequences of predator-tadpole interactions on the rest of the aquatic communities of temporary ponds, but amphibians' impact on the community structure and dynamics of aquatic systems may be conditioned by their interactions with competitors and predators. Thus, predators can potentially have cascading effects on pond communities via alterations in amphibian larval density and/or their phenotype. We used a mesocosm array at Doñana National Park to test for density dependent and density-independent effects of native and invasive predators on survival and growth rate of amphibian larvae, also quantifying its consequences on the food web. Invasive predators caused greater mortalities of amphibian larvae than native predators, hence having a stronger potential for direct density-dependent effects on amphibian guilds and their interactions in the trophic web. Moreover, cravfish increased turbidity and nutrient content in the water and had a direct negative impact on plants that was carried over into the following hydrological cycle. Thus, our results highlight how this invasive species poses a greater predatory threat to amphibians than other native predators, and can also be highly disruptive of the entire pond structure.

D23SY31PS0268

NUTRITIONAL ECOLOGY AND CONTEST BEHAVIOUR IN PARASITOID WASPS

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Animal contests are widely studied by behavioural ecologists and there is a well-developed framework for evaluating their functional significance. Parasitoid wasps in the family Bethylidae (Insecta: Hymenoptera) are ideal organisms to test evolutionary models as all of the predicted major influences on contest outcome-fighting ability, prior ownership and the way that individuals value possession of the contested resource - have now been successfully explored. However, possible links between nutritional/physiological state and contest behaviour have not been investigated. Parasitoids can feed on nutritionally very different diets during stages of their life cycle, such as nectar, honeydew and hosts. It is expected that particular nutrients may affect contest ability (Resource Holding Potential, RHP), whereas others may influence the value of the resource (RV) to a contestant. Here we explore whether dietary carbohydrates influence RHP and/or RV and, consequently, contest resolution. The influence of nutritional state on contest behaviour is evaluated with behavioural observations of dyadic contests of parasitoids wasps, whereas the Identification and manipulation of key dietary nutrients is achieved by metabolomics analysis (LC-MS and proton NMR spectroscopy).

D23SY31PS0315

CONFLICT AND CO-ADAPTATION: THE EVOLUTION OF PARENTAL CARE IN A WILD BIRD SPECIES

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Interactions between caring parents and their offspring are one of the most widespread social behaviours in animals. Parent-offspring interactions involve two parties and its evolution is, therefore, dependent on the evolution of two traits, parental provisioning and offspring begging. From a quantitative genetic perspective both behaviours should co-adapt, which should ultimately lead to (genetic) co-variation. This hypothesis is supported by a number of recent empirical studies. However, most of these studies have been performed in captivity under controlled environmental conditions. This limits the information on among others fitness costs, while these costs are thought to stabilize the process of co-adaptation. By means of a cross-fostering experiment, we studied co-adaptation of offspring begging and parental provisioning in free-living blue tits (*Cyanistes caeruleus*), thus in the presence of natural selection. We did find evidence for co-adaptation, but only in male parents, as the paternal feeding rates co-varied with begging intensities of their genetic chicks, which were raised in foster nests. By contrast, feeding rates of females co-varied with the begging intensity of the foster chicks they were raising. We can think of two hypotheses that may explain the observed pattern: First, the co-variation in males may reflect a (general) genetic correlation between parental feeding rates and offspring begging, which may be masked in females due to a higher responsiveness to offspring need (thus foster chick begging). A higher responsiveness to actual need in females may also indicate that they may compensate for deviations between offspring begging and paternal feeding. Second, the covariation of fathers' feeding rate with their genetic offspring's begging may relate to prenatal maternal effects, with mothers interlinking offspring begging (possibly via differently allocation of egg compounds) to the males' expected contribution to parental care.

D23SY31PS0343

MATING SUCCESS AND MORPHOLOGICAL TRAITS IN DROSOPHILA MELANOGASTER STRAINS DEVELOPED IN DIFFERENT NUTRITIVE ENVIRONMENTS

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Relations between mating success, adult body size and correlated traits were among the more explored in sexual selection studies. Reproductive behavior of many dipteran species, including Drosophila, depends of larval nutrition by influencing various aspects of adult body size. Nutrient-induced differences in traits that represent body size may affect both sexes, through competition (in males) or affecting fecundity (in females). Then, it is important to establish how fruitflies perceive the complex chemical environment, i. e. different chemical compounds involved in their life activities, such as feeding, mating and oviposition. In a series of multiple choice experiments, *Drosophila melanogaster* flies were put in competitive mating scenario and scored for mating success. *Drosophila melanogaster* strains maintained for more than a decade on standard laboratory medium and substrates containing fruits or vegetables were used. After behavioral observations, flies were used in analysis of morphological traits such as wings and sex combs. Wings are involved in male courting activities, due to attractiveness in their morphology (size, shape) and production of acoustic stimuli. Sex combs, which represent male secondary sexual trait, are also included in exchange stimuli during courtship. Beside their potential role in mating behavior, both wings and sex combs may be useful traits for considerations of effects of nutritional environment. Flies reared on different diets showed differences in mating success. As wing size and shape sexual dimorphisms were observed, the effects of long-term exposure to different nutrition on morphological characteristics under this study will be also considered. Obtained results will be further discussed in light of relations established among nutrition, behavioral and morphological traits in this species.

D23SY31PS0344

QUANTITATIVE TRAIT GENES FOR FEARFUL BEHAVIOUR UNDER CHICKEN DOMESTICATION

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Domestication is a form of strong directional selection imparting wide-ranging phenotypic changes to animals and plants, and ever since Darwin considered a model of evolution. Changes in behaviour, particularly fearful and social behaviour, are at the heart of animal domestication. A wild by domestic advanced intercross of chickens is a powerful study system for the genetics and genomics of domestication phenotypes. We applied quantitative trait locus (QTL) mapping and genetical genomics to fear-related behaviours. QTL mapping in 572 birds from an eight-generation intercross revealed ~44 loci for behaviour in three test situations: an open field, a social reinstatement, and a tonic immobility test. The tests have separate but overlapping architectures with a few potentially pleiotropic loci and small to moderate QTL effect sizes. To search for underlying genes we mapped transcriptome-wide expression QTL (eQTL) in hypothalamus from 129 birds. Out of 634 eQTL, 16 candidate quantitative trait genes had eQTL coinciding with behaviour QTL and a gene expression—behaviour correlation. Structural equations modelling found eight genes in four QTL to be consistent with a causal role of gene expression: *PRDX4* (a periredoxin), *ACOT9* (an acyl-coenzyme A thioesterase) and *SRPX* (Sushi repeat-containing) are candidates for a social reinstatement and tonic immobility locus on chromosome 1; TTRAP (TRAF and TNF receptor-associated protein) and an unknown EST sequence 60386624F1 for a social reinstatement QTL on chromosome 2; ADAM10 (disintegrin and metalloproteinase domaincontaining) and APBA2 (Amyloid beta A4 precursor protein-binding) for an open field locus on chromosome 10; and the unknown LOC770352 for a second open field QTL on chromsome 10. In conclusion, our mapping gives genetic and gene expression evidence for unexpected putative quantitative trait genes for fearful behaviour under chicken domestication.

D23SY31PS0361

NEST DISTRIBUTION AFFECTS BEHAVIOUR AND REPRODUCTIVE SUCCESS OF A MARINE FISH

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The distribution of breeding resources, such as nest sites, can have a pronounced impact on a population by affecting the proportion of individuals that succeed to breed and hence the variation in reproductive success. Aggregation of important resources can lead to resource monopolisation by a limited number of individuals, and thus affect the intensity of sexual selection. We tested by contrasting two experimental treatments (dispersed vs. aggregated) how nest distribution affects: (1) mating behaviour, (2) male nest occupation and mating success, and (3) reproductive success and the opportunity for selection. We used the two-spotted goby (Gobiusculus flavescens), a small marine fish with a resource-based mating system, as our model species. When nests were aggregated, a larger proportion of the males behaved aggressively, fewer males succeeded in occupying a nest, fewer males became mated, and those males that mated received fewer eggs from spawning females. These effects resulted in a higher variance in reproductive success, and hence a higher opportunity for selection (Irs), in the aggregated treatment. However, we found no significant selection differentials for body length or condition of males in either treatment. Our results support the hypothesis that aggregation of essential resources like nests promotes resource monopolisation. In species facing highly clumped nesting resources in the wild, monopolisation may negatively impact population productivity but could lead to strong selection on traits that promote male competitive ability.

D23SY31PS0371

MOLECULAR CHARACTERIZATION OF POSTZYGOTIC ISOLATION BARRIERS IN WILD TOMATOES: THE ENDOSPERM TRANSCRIPTOME OF SOLANUM PERUVIANUM

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The establishment of reproductive isolation barriers is a basic component of the speciation process and thus of major interest in evolutionary biology. Closely related species in the tomato clade (Solanum section Lycopersicon) provide interesting material to study both pre- and postzygotic barriers. The sister species Solanum peruvianum and Solanum chilense exhibit very strong, reciprocal postzygotic reproductive isolation; following controlled pollinations there is production of fruits but developing embryos are usually aborted, i.e. the seeds are inviable. Embryo inviability (hybrid seed failure) is a common interspecific barrier in angiosperms, and there is substantial evidence that imbalances in endosperm-embryo interactions early in seed development mediate such barriers. Our histological comparisons of hybrid and intraspecific seeds of these species have shown abnormalities in hybrid endosperm development that are concurrent with arrest in embryo growth. We hypothesize that key genes involved in endosperm development, as well as epigenetic factors mediating their expression (i.e. small RNAs and genomic imprinting), are disrupted in these hybrid crosses. Following reciprocal within-population crosses, we laser-microdissected endosperms of early-developing seeds of *S*. peruvianum, extracted RNA and sequenced transcripts on the Illumina HiSeq2000 platform. The composition of the "normal" intraspecific endosperm transcriptome and its expression levels will be characterized. The next steps will be an assessment of the hybrid endosperm transcriptome (following reciprocal crosses between *S. peruvianum* and *S. chilense*) and its comparison with the within-species transcriptome. This research shows how endosperm development and its abnormalities in hybrid seeds are an important force shaping the speciation process in the tomato clade.

D23SY31PS0482

SMALL RNA REGULATION OF POST MATING RESPONSE TO SEX PEPTIDE IN FEMALE DROSOPHILA MELANOGASTER

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Male *Drosophila melanogaster* fruitflies transfer seminal fluid proteins (sfps) to females during mating. One sfp, sex peptide, is responsible for several post mating responses in females, such as increased egg laying and reduced remating rate. Yet little is known about the female responses to these substances at the genomic level. Small RNAs are short (20-22nt) regulatory non-coding RNAs, which bind to messenger RNAs and prevent their translation to proteins. By the use of 'high definition' adapters, we reduced the bias in small RNA sequencing data detection and measurement of small RNAs. We investigated here the differences in expression of small RNAs and messenger RNA in female *D. melanogaster* mated to males that do and don't transfer sex peptide during mating. We assayed different tissue types at four consecutive, equally distributed time points after mating. The results show how gene regulation following receipt of sex peptide is influenced by small RNAs. We discuss the results in the context of sexual conflict and post mating responses in females.

D23SY31PS0498

WHY DO SPIDER MITES RE-MATE?

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In Tetranychus urticae, only the first mating is effective, except if the interval between first and second copulations is shorter than 24 hours or if the first mating is interrupted. However, males often attempt to copulate with mated females. Here, we address this paradox. We first tested whether males prefer to mate with females that have mated within the preceding 24 hours or with those that have mated before that period. We found that males show no preference between these two types of females. Moreover, the time to mating was longer and the mating duration shorter when males mated with mated females, relative to virgins, irrespective of their timing of mating. This confirms a lack of distinction between different types of mated females and suggests that males are either less motivated to mate with mated females or that the second mating occurs for a different reason than siring offspring. Subsequently, we investigated the consequences of polyandry for the reproductive fitness of females, depending on the frequency and timing of the mating events. We predicted that if females benefit from polyandry, fecundity and survival of multiply mated females would be higher than those of once mated females. Indeed, multiply mated females have higher fecundity than once mated females, suggesting that females potentially benefit from mating multiply. No difference in survival and sex ratio was found between these females. Our data shows that females benefit from multiple matings, hence this behaviour is probably under female control. This result has implications for our understanding of mating behaviour in spider mites and other organisms.

D23SY31PS0561

SEXUAL SELECTION ON A CRYPTIC WING PATTERN TRAIT IN DROSOPHILA

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Wing interference patterns (WIPs) are a newly discovered trait in *Drosophila*, and are caused by thinfilm interference of light on the wing surfaces. Only 10-12% of the incident light on the wing is reflected as a WIP, so these patterns are only visible against a dark background. This is also the reason WIPs are a previously overlooked (i.e. cryptic) trait. We have carried out the first comprehensive study of WIPs in *Drosophila*, and have been able to confirm that although WIPs are influenced by wing size, there is a large genetic component to WIP variation. We have also carried out a manipulative mate choice experiment, where female preference for different WIP phenotypes was tested against light and dark backgrounds. We found that there was a significant effect of WIPs on female preference in the dark background, providing the first evidence that WIPs are a cryptic sexually selected trait in *Drosophila melanogaster*.

D23SY31PS0608

VULTURES ACQUIRE INFORMATION ON CARCASS LOCATION FROM SCAVENGING EAGLES – THE EVOLUTION OF A PRODUCER-SCROUNGER GAME

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Vultures are known as the scroungers of the natural world, owing to their ecological role as obligate scavengers. In addition to being unable to kill prey for themselves we suggest that they are dependent on other species to locate food. Our analyses of bird arrival times at carcasses show that vultures join a carcass after discovery by a raptor more often than expected by chance. We therefore hypothesise that a producer-scrounger game is at play, with raptors tending to locate and "produce" carcasses in an information sense, while the vultures tend to "scrounge" this information by taking their lead from the raptors. We develop a game-theoretic model showing that such an interspecific producer-scrounger game is evolutionary stable when vultures are dominant to the raptors in agonistic interactions over food. Based on this theoretical finding we explored the dominance hierarchy at a carcass and show empirically that vultures dominate raptors in aggressive contests over food. Together our analyses show that vultures scrounger game. In terms of information use, raptors are dependent on personal information whereas vultures use socially acquired information. Globally vulture populations are declining so our findings have implications for the conservation of the group. By sometimes depending on raptors to locate carrion, scrounging vultures are sensitive to declines in the abundance of these producers.

D23SY31PS0722

DAILY AND SEASONAL CHANGES IN THE LOCOMOTOR ACTIVITY OF DROSOPHILA MONTANA FLIES IS ACCOMPANIED BY CHANGES IN THE EXPRESSION OF TWO CIRCADIAN GENES, PERIOD AND TIMELESS

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Adaptation to daily and seasonal changes in environmental conditions is of crucial importance for the species living in northern latitudes. This includes the ability of the organisms to predict the forthcoming winter by sensing gradual changes in environmental cues early enough to be prepared e.g. by synchronizing their metabolic, developmental and behavioral processes along with the changing seasons. This forecasting system relies heavily on the function of two major molecular clock mechanisms, the circadian clock and the photoperiodic timer. The genetic and neuronal background of the circadian clock is well-known, but the possible role of circadian clock in the photoperiodic timer is under intense debate, partly due to the fact that most of the studies have been performed in D. melanogaster that is not adapted to live seasonally highly varying environments and possesses only mild seasonal photoperiodic responses. Our study species, D. montana, offers a unique opportunity to study these questions as this northern species shows clear circadian rhythms as well as robust photoperiodic responses. We have studied the possible involvement of the circadian clock in seasonal adaptation by tracing the daily and seasonal rhythms in the locomotor activity of D. montana females and the function of two circadian clock genes, period and timeless, in conditions mimicking light and temperature conditions during 'summer' and 'autumn', when the females' developmental pathway (direct maturation vs. reproductive diapause) is determined. D. montana flies shifted their evening activity, as well as the expression peaks of per and tim, towards an earlier time of the day along with a decrease in day length, suggesting that these genes are involved in seasonal adaptation. In addition, a decrease in D. montana's locomotor activity, as well as the lower activity of diapausing females vs. the non-diapausing ones in late summer and autumn are clearly adaptive features on northern latitudes.

D23SY31PS0740

QUANTIFYING GENE EXPRESSION PATTERNS FOR UNDERSTANDING DEVELOPMENT AND EVOLUTION

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To understand morphogenesis and the evolution of organs it is crucial to visualize and quantify gene expression patterns. Development of high-throughput imaging techniques, such as optical projection tomography (OPT), to visualize gene expression patterns in a 3D framework has revolutionized developmental biology research. However, methods to precisely quantify gene expression domains in a systematic manner are lacking. To fill in this gap our research focuses on developing novel methods for multivariate shape analysis and comparison of gene expression domains within developing structures. Here we used the limb bud as a model for development and analyzed the gene expression domains of two relevant genes for proximo-distal limb patterning, Hoxa11 and Hoxa13, in a large sample of C57Bl6 mouse embryos between stages E10-E12. Gene expression domains were segmented from whole-mount in situ hybridization experiments with an automatic multiple thresholding method. Gene expression patterns were compared across stages and in association with limb morphology using geometric morphometrics. Our method provides an accurate phenotyping tool to quantify the spatiotemporal dynamics of gene expression patterns. Multiple thresholding enables to capture the continuous gradient of gene expression domains by overlapping the isosurfaces thresholded for different values of gene expression. Comparison within and between mouse litters at different timepoints highlights the complexity and variation of gene activity underlying limb morphogenesis to an unprecedented level of detail. Further development of methods to compare gene expression patterns in mutant versus wildtype strains and between different species may increase our ability to track development and to investigate how evolution has modified a common developmental pattern to generate a wide diversity of morphologies, as in the vertebrate limb. Potentially, these methods could be extended to different organs, genes and animal models.

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CHARACTERIZATION OF SEX-BIASED GENES FROM THE TRANSCRIPTOME OF A MALE-DIMORPHIC MITE

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The different sexes experience different selective pressures, which can lead to highly divergent phenotypes that are achieved via sex-biased gene expression. As a result of sexual selection and sexual conflict, sex-biased genes are expected to evolve at a faster rate than other genes in the genome. Furthermore, because sexual selection acts more strongly on males, male-biased genes are expected to evolve faster than female-biased genes. We aimed to test these predictions in a model species for sexual selection and conflict research, the bulb mite Rhizoglyphus robini (Acari, Acaridae), in which armored, aggressive fighter males coexist with unarmored scrambler males. We sequenced and de-novo assembled transcriptomes of adult males and females. Expression of 4.0% of 114,456 transcriptomebased gene models (TGMs) was male-biased, whereas 1.3% of TGMs had female-biased expression and this difference was highly significant. For TGMs with protein-coding capacity, the proportion with orthologs identified in the spider mite (Tetranychus urticae) genome was lower for male-biased than for unbiased genes, but was very similar for female-biased and unbiased genes. Mean amino acid distances between male-biased R. robini genes and their T. urticae orthologs were significantly greater than those between unbiased genes and their orthologs; female-biased gene orthologs had distances similar to those of the unbiased orthologs. The higher number and faster evolution of male-biased genes are consistent with the hypothesis that sexual dimorphism is driven mostly by male-biased sexual selection. This hypothesis is further supported by the result that the fighter morph, which is behaviorally and morphologically adapted to intense intrasexual competition, was characterized by a considerably larger number of overexpressed (compared to female expression levels) genes than the less sexually dimorphic scrambler morph.

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POSITIVE SELECTION DRIVES THE TRANSCRIPTIONAL EVOLUTION OF THE DROSOPHILA CHEMOSENSORY GENE FAMILIES

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The chemosensory system is involved in critical processes such as nutrition, reproduction and social communication, being therefore crucial for the survival of animals. In insects, the early steps of the chemosensory process are mediated by proteins encoded in multigene families, including extracellular ligand-binding proteins (OBPs, CSPs and CheBs) and membrane receptor proteins (ORs, GRs and IRs). Since the ability to discriminate chemical stimuli (and thus the individual fitness) depends on such gene families, positive selection may promote their transcriptional evolution as an adaptation to external environment changes. Here we examined the transcriptional changes underlying the adaptive evolution of the chemosensory gene families. In particular, we analysed the OBP gene cluster organisation, the turnover of cis-regulatory elements, and the patterns of polymorphism and divergence at the upstream regions of the genes of these chemosensory families. For that, we integrated information from diverse genome-wide data sets, such as the expression pattern, chromatin state and promoter architecture of the confined genes. We found that chromatin domains play an important role in the maintenance of the OBP clusters by restricting the location of genes to regions with the appropriate transcriptional environment: low expression intensity, high expression breadth and noise. These elevated noise levels may be adaptive, by increasing the behavioural plasticity in front external changing environments. Moreover, we also identified the positive selection signature at the nucleotide variability of the upstream chemosensory regions, as well as in the turnover of cis-regulatory elements. Therefore, our results show that positive selection has driven the transcriptional evolution of the chemosensory gene families.

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A RESAMPLING-BASED APPROACH TO STUDY VARIATION IN MORPHOLOGICAL MODULARITY

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Modularity has been suggested to be connected to evolvability because an higher degree of independence among parts allows them to evolve as separate units. Recently, the Escoufier RV coefficient has been proposed as a measure of morphological modularity in multivariate morphometric datasets. However, it has been shown, using randomly simulated datasets, that the value of the RV coefficient depends on sample size. Also, so far there is no statistical test for the difference in modularity between *a priori* defined groups. Here we: 1. using a rarefaction analysis, show that the value of the RV Escoufier coefficient depends on sample size also in real morphometric datasets; 2. propose a permutation procedure to test for the difference in the RV coefficient between *a priori* defined groups; 3. show, through simulations, that such a permutation procedure has an appropriate Type I error. The permutation procedure outlined here, readily extendable to non-morphometric datasets, will allow statistically sound comparisons of the degree of modularity between *a priori* defined groups.

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SEX-BIASED GENE EXPRESSION IN A BROADCAST SPAWNING DIOECIOUS BROWN ALGAE, FUCUS VESICULOSUS

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The fucoid brown algae are increasingly the focus of ecological genetics, biodiversity, biogeography and speciation research. The molecular underpinnings of mating system variation (repeated dioecious – hermaphrodite switches) and the molecular evolution of sex-related genes, as well as proteins involved in gamete interactions are hampered by a lack of genomic information. We undertook a comparative analysis of male and female reproductive tissue transcriptomes (FLX 454) against a vegetative background during natural reproductive cycles in *Fucus vesiculosus*. Over 300k reads were assembled and annotated against public protein databases. Photosynthetic and carbohydrate metabolism pathways were under-expressed, particularly in male tissue, while several pathways involved in genetic information processing and replication were over-expressed. Estimates of sex-biased gene (SBG) expression were higher for male (14% of annotated orthologues) than female tissue (9%) relative to the vegetative background. Mean expression levels and variance were also greater in male- than femalebiased genes. Major female SBG were carbohydrate-modifying enzymes with likely roles in zygote cell wall biogenesis and/or modification. Male SBG reflected distinct sperm development and function, and orthologues for signal perception, transduction, and putatively flagella-localized proteins. Overall, the results suggest constraint on female SBG (possible pleiotropy), and less constrained male SBG associated with sperm-specific functions. Our results support the growing contention that males possess a large array of genes regulating male fitness, broadly supporting findings in evolutionarily distant heterogametic animal models. This work identifies an annotated set of *F. vesiculosus* gene products that potentially regulate sexual reproduction and contribute to prezygotic isolation, one essential step towards developing tools for a functional understanding of species isolation and differentiation.

D23SY31PS0930

A NOVEL APPROACH TO STUDY PHENOTYPIC EVOLUTION IN AN ADAPTIVE RADIATION: X-RAY IMAGES PROVIDE NOVEL INSIGHTS INTO THE ADAPTIVE RADIATION OF CICHLID FISHES IN EAST AFRICAN LAKE TANGANYIKA

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The East African Great Lakes, Tanganyika, Victoria and Malawi, are ecosystems with highly diverse habitats and food resources and renowned for the spectacular adaptive radiations of cichlid fishes that they contain. Lake Tanganyika harbors a highly diverse cichlid species flock comprising more than 220 species that are morphologically and ecologically highly diverse, but also show a number of convergent cichlid species that have evolved within the lake. Here, we use a novel approach to study convergence and divergence in Lake Tanganyika cichlids, making use of about 5000 x-ray images of more than 160 Tanganyikan cichlid species. We used geometric morphometric as well as meristic measurements in combination with ecological data (stable-isotopes) and a molecular phylogeny to examine the patterns of diversification in a large cichlid radiation. Furthermore, we were interested in testing predictions regarding adaptive radiation, such as its progress in stages, the early-burst scenario and the correlation between phenotypes and the environment. We identified various instances of convergent evolution within the adaptive radiation of cichlid fishes in Lake Tanganyika, particularly regarding body elongation and benthic or pelagic lifestyles and show a relatively constant rate in phenotypic evolution. Our study thus deepens the understanding of how cichlids diversify and how morphologies changed as species adapted to different habitats and foraging modes.

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THE COSTS OF HELPING BEHAVIOUR IN COOPERATIVELY BREEDING WARBLERS

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In several animal species, subordinate individuals forego independent breeding to assist others in raising offspring that are not their own. Several benefits underlying the evolutionary stability of such seemingly altruistic helping behaviour have been invoked, and assessment of variation in individual investment in response to variation in such benefits is often being used to reveal the adaptive significance of these benefits. In the Seychelles warbler Acrocephalus sechellensis, subordinates gain substantial fitness benefits of helping: individuals that start as helpers have higher life time fitness than subordinates that do not help initially. Nonetheless only about half of the subordinates do help. Using a range of physiological measures, we show here that helping carries a substantial cost, as helpers were in poorer condition after the breeding season than non-helping subordinates . Additionally, only individuals in good condition do provide help, because probably only these individuals can overcome the associated costs. This result suggests that, apart from the associated benefits, variation in subordinates' condition explains part of the variation in helping are important to unravel the benefits of helping, and to understand the variation in the expression of helping behaviour within species.

D23SY31PS0977

ALTERATION OF HPA/HPG AXIS REACTIVITY AS A CONSEQUENCE OF CHICKEN DOMESTICATION

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Domesticated animals can serve as models to study evolutionary processes. The hormonal stress responses of wild and domesticated animals have rarely been thoroughly compared. The Red Jungle fowl (RJF) is considered to be the main ancestor of all domesticated chicken breeds. The aim of this study was to investigate the impact of domestication on behaviour and reactivity of HPA/HPG axis to a stressful physical restraint episode. Using liquid chromatography tandem mass spectrometry methods (LC-MS/MS), plasma concentrations of 5 classes of steroids, namely, pregnanes, progestines, androgens, estrogens and glucocorticoids were measured at basal level, 10 and 60 minutes after restraint in domesticated female White Leghorn (WL) and RJF. In behaviour tests, WL had a slower stress recovery, whilst RJF resumed baselines in behaviour more quickly. Corticosterone level was significantly influenced by the stressor ($p \le 0.001$) in both breeds. RJF had significantly higher acute stress response (p ≤ 0.05) but quicker stress recovery compared to WL. In RJF, most other hormones were not influenced by the stressor while they were mostly significantly affected in the domesticated WL. Among the hormones which were differently altered in WL and RJF, dehydroepiandrosterone (DHEA), an androgen which is also involved in social and aggressive behaviour in birds, showed the most pronounced breed difference ($p \le 0.005$) and response to stress ($p \le 0.001$) in WL and might be highlighted as an important hormone in relation to both stress and domestication of chicken. To our knowledge this is the first comprehensive study, investigating the impact of stress on behaviour and a wide range of steroid and neurosteroid hormones in any domesticated animal and it's wild ancestor. In conclusion, our study shows that the domesticated phenotype in chicken is related to lower HPA axis reactivity and higher HPG axis activity, which cannot be maintained in stressful situations.

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EFFECTS OF A MUTATION IN THE ADRENERGIC RECEPTOR GENE ADRA2C, SELECTED DURING CHICKEN DOMESTICATION, ON GENE EXPRESSION, RECEPTOR DENSITY AND BEHAVIOUR

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During domestication, chickens have been selected for genotypes which confer selective advantages in conditions under human care and rearing, but little is known about the detailed genetic mechanisms involved. In a recent study of selective sweeps associated with domestication, resequencing of different chicken breeds reveled a number of strongly selected loci. One of those, containing the α -adrenergic receptor 2C (ADRA2C), was found to be selected in layer chickens. Furthermore, previous studies from our group show that the receptor is hyper-methylated in White Leghorn (WL) chickens compared to the Red Junglefowl (RJF) ancestor, suggesting the gene to be differently expressed. ADRA2C is a presynaptic autoreceptor modulating epinephrine and norepinephrine release within the central nervous system and secretion of epinephrine from the adrenals. A down-regulation is associated with increased catecholamine secretion. Hence, domestication on the receptor might have an impact on stress responses, memory formation and egg production. We used F9-birds from an intercross between domesticated WL and RJF, and selected parents which were heterozygous for the mutation in ADRA2C generating offspring with all genotypes represented. This allowed within-family comparisons of the effects of the mutation against a randomly recombined genome. We used an array of stress related behavioral tests and we measured differences in brain expression of the gene as well as receptor density. Briefly birds with the mutation tended to have impaired associative learning and recovered more quickly after exposed to an aerial predator model. We hypothesize that the domestication of chickens have selected birds with hypo-expressed levels of ADRA2C, generating an increased catecholamine release, and that this has caused modifications of the stress response.

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EXPLORATORY PERFORMANCE IN AN OPEN FIELD TEST OF BANK VOLES (MYODES GLAREOLUS) FROM MULTIDIRECTIONAL ARTIFICIAL SELECTION EXPERIMENT

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It has been proposed that evolution of complex physiological adaptations, such as endothermy or capability to utilize a specific diet, could be driven by natural selection acting primarily on behaviour. Behavioural trait such as "personality" of individuals, influences habitat preference, aggression, attitude to novel objects or situations, which in turn could lead to the variation in physiological traits such as metabolic rate. As a consequence, individuals with higher resting metabolic rate are able to collect, process, and invest more energy, complementing their active personality. However, empirical evidence for this hypothesis remains ambiguous. To test the hypothesis we are comparing exploratory behaviour of bank voles (*Myodes glareolus*), selected for high aerobic capacity (A), predatory aggression (P), ability to grow on low quality herbivorous diet (H), with that of unselected control (C), in an open field condition. By measuring exploratory performance in an environment like open field, which is novel and stressful to these semi-fossorial rodents, we would interpret personality of individuals across the selection lines, on an "proactive – reactive" axis. We hypothesize that voles from A and P lines will show more "proactive", whereas those from H lines more "reactive" personality, compared to voles from unselected C lines. The results will enable us to investigate if variation in personality is heritable and genetically correlated with the traits under selection.

D23SY31PS0993

GENETIC INSIGHTS INTO THE BEHAVIOURAL AND MORPHOLOGICAL DIVERSITY OF AFRICAN MOLE-RATS (FAMILY: BATHYERGIDAE)

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A principal constraint to have shaped many aspects of African mole-rat biology is their strictly subterranean lifestyle, with individuals rarely emerging above-ground. Linked to this lifestyle, their diet consists mostly of geophytes, the storage organs of plants, which has allowed mole-rats to occupy a range of climatically varied habitats across sub-Saharan Africa. Morphological adaptations include an elongated cylindrical body with shortened limbs, reduced eyes and external ears, and even hairlessness in the naked mole-rat (Heterocephalus glaber). Physiologically, mole-rats are adapted to a life in constant darkness, which may affect their circadian rhythms, and reduced oxygen levels (hypoxia), which may have allowed them to become insensitive to certain pain stimuli. Furthermore, for rodents, mole-rats are exceptionally long lived (~30 years). Social structure across the family ranges from solitary to eusociality, with sociality thought to have evolved multiple times. A subterranean lifestyle and similar social structures have convergently evolved in other divergent families within the Rodentia. All of these facts, coupled with the resource of the recently completed naked mole-rat genome, make this family a valuable group for diverse evolutionary studies. To identify genes that have been targets of selection in this group, we are undertaking phylogenomic analyses of mole-rats using transcriptomes obtained from 8 ecologically diverse mole-rat species, including social and solitary species. We present our preliminary findings of comparative genetic analyses of candidate genes relating to some of the key adaptive traits, for example, those relating to sensory perception, locomotion and social behaviour, which have evolved within this family. With the aim of beginning to document and understand the genetic basis behind the numerous remarkable evolutionary adaptations shown by the members of this family of highly specialised hystricomorph rodents.

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GENE EXPRESSION DIFFERENCES IN ARCTIC CHARR ECOMORPHS DURING HEAD DEVELOPMENT

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There is a strong connection between microevolutionary and ecological processes during the emergence of intraspecific diversity. Within populations similar phenotypes can be found in similar environments and hence appear as intraspecific parallel evolution. A remarkable example of this is the repeated occurrence of sympatric morph pairs (benthic/limnetic) in northern freshwater fishes. Arctic charr (Salvelinus alpinus) exhibits parallel evolution of benthic/limnetic morphological characteristics, particularly in the head and trophic apparatus. In Lake Thingvallavatn, Iceland, four ecomorphs of Arctic charr are found, two are benthic with subterminal mouth and blunt head (small and large benthivorous) and two are limnetic with elongate gracile terminal mouth (planktivorous and piscivorous). In this study the molecular basis of this craniofacial divergence was addressed during embryonic development by spatio-temporal expression profiling of candidate genes using a combination of tools (i.e. RNA-Seq technology, Real-Time QPCR, and in situ hybridization). Among differentially expressed candidate genes, a cluster of genes with craniofacial specific expression pattern have shown significant upregulation in the head of benthic ecomorphs. Two members of matrix metalloproteinase family, MMP-2 and MMP-9, were amongst the top differentially regulated candidates. Interestingly, a previous study in zebrafish demonstrates that glucocorticoid-induced increase in expression and activity of MMP-2 and -9 during embryogenesis generates a paedomorphic phenotype with subterminal mouth similar to benthic charr morphology. Thus, we hypothesize that a mechanism for the differential regulation of MMP-2 and -9 may play an important role in determining the benthic/limnetic craniofacial divergence of Arctic charr ecomorphs in this lake.

D23SY31PS1087

CO-OPTION OF HOST-ALTERNATING POLYPHENISM IN THE EVOLUTION OF CASTE POLYPHENISM IN SOCIAL APHIDS

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Castes in social insects are one of the most well-known examples of polyphenism. However, the developmental origins of the discrete alternative phenotypes are largely unknown. Social aphids form galls on their primary host plant and open colonies on their secondary host plant, producing different phenotypes on each plant. Aphids of the genus Colophina form galls on Zelkova trees in spring. Winged aphids migrate from mature galls in summer and then found open colonies on *Clematis* plants. In the galls, polyphenism does not occur and the aphids produce first-instar defenders that attack predators with mouthparts. In the open colonies, first-instar sterile soldiers with enlarged legs and shortened mouthparts are produced from polyphenic development. The morphology and defensive behaviour of the soldiers in the open colonies resembles those of monomorphic defenders in the galls, suggesting the possibility that these two defensive morphs share a common developmental program. To investigate the genetic relationship between defenders of Zelkova trees and soldiers of Clematis colonies, I compared morphological traits of four Colophina species. Body parts of the following morphs were measured: 1. monomorphic first-instar nymphs of Zelkova galls, 2. reproductive firstinstar nymphs of *Clematis* colonies, 3. soldier first-instar nymphs of *Clematis* colonies. Morphometric analyses showed a positive correlation between the morphology of the first-instar nymphs in galls and the sterile soldiers in open colonies. Moreover, in the species close to their ancestral condition, both phenotypes were almost identical. These results suggest that the soldier caste of *Colophina* species first evolved by co-option from ancestral host-alternating polyphenism, and diverged to have specialized morphology. I will also discuss ecological factors favouring the evolution of the sterile castes, and the contribution of inclusive fitness to the evolution of a novel phenotype.

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EXPERIMENTAL EVOLUTION REVEALS THE "MOTHER'S CURSE": SPERM COMPETITION AND CYTONUCLEAR INTERACTIONS IN DROSOPHILA

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Matrilineal transmission creates the potential for male-specific deleterious variation to accumulate in the mitochondrial genome via either selection for female-benefit sexually antagonistic alleles or drift. In either case, the result may be increased selection for nuclear regulation of mitochondrial function in males. Sperm function may therefore be particularly sensitive to cyto-nuclear coadaptation and compensatory evolution. Mitochondria are known to play a key role in insect spermatogenesis, though the energetics of insect sperm are relatively poorly understood. To investigate whether cyto-nuclear interactions are important in the evolution of sperm competitive ability, we created cyto-nuclear hybrids between Drosophila melanogaster populations selected for different life histories. We used five pairs of experimentally-evolved populations, now divergent for over 1000 generations. One selection treatment, CO, has been selected for late life reproduction (4 wk cycle). The other, ACO, has undergone selection for extremely rapid development and very early reproduction (9 day cycle), leading to marked changes across a host of life history and morphological traits (the "fast development syndrome"). We measured sperm offensive ability (P2) of all 20 cyto-nuclear hybrid and purebred populations at early and late ages by double-mating control females with a standard competitor as the P1 male and recording siring success. ACO-nuclear males with CO cytoplasm had lower P2 success than pure ACO males at both ages, while CO-nuclear males with ACO cytoplasm had lower success than pure CO males in old age. The deleterious effect of foreign cytoplasm on sperm competitive function suggests cyto-nuclear interaction can play an important role in the evolution of spermatogenesis or sperm energetics; which of these factors has evolved is the object of current investigation.

D23SY31PS1144

TSHR - A POSSIBLE DOMESTICATION GENE

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Domestication, the process where wild animals adapt to captivity, offers an excellent model to study evolution. Strong selection for traits of human interest causes genetic adaptations and a rapid change in morphology, physiology and behaviour. The Red Jungle Fowl (Gallus gallus) is the wild ancestor of domesticated chickens. Resequencing has revealed a selective sweep in domestic chickens over the thyroid stimulating hormone receptor gene (TSHR). A missense mutation in TSHR causing a glycine to arginine change is the most obvious candidate causal mutation for the sweep. Thyroid hormones are important in development and the thyroid system is suggested to correlate with stress response and hence behaviour in animals. Fast development and altered behavioural responses are traits shared by all domesticated animals. Therefore, we hypothesize that a mutation in the TSHR gene has been of selective advantage during the domestication of the chicken. To study this, we used intercrosses between Red Junglefowl and domesticated White Leghorns. We included 28 birds homozygous for the TSHR mutation, 34 homozygous for the wild-type allele and 62 heterozygous birds, and subjected them to a broad phenotyping. Incubation time was recorded in order to measure development and behavioral tests for social, aggressive and fear behaviors were conducted. Individuals homozygous for the TSHR mutation had longer incubation time $(21,7 \pm 1,3 \text{ vs. } 18,0\pm 1,2 \text{ and } 18,2\pm 1,0 \text{ h}$ from first hatch p<0,05), showed less aggression in a social dominance test (8,6±4,8 vs. 26,9±4,8 and 18,6±4,8 number of observations p<0,05) and less fear in a fear of human test (2,2±2,5 vs. 14,9±2,3 and 16,3±1,9 % of observations p<0,05) in comparison to the other genotypes. The results are in line with previous studies comparing pure-bred domestic White Leghorns with pure-bred Red Jungle Fowl. This indicates that a mutation in the TSHR gene could have been of selective advantage during the domestication of the chicken.

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THE INFLUENCE OF CYTO-NUCLEAR INTERACTIONS ON LIFE-HISTORY TRAITS AND BEHAVIOUR

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Despite the longstanding perception of the selective neutrality of mitochondrial genes, there is a growing awareness of their influence on life-history traits via interaction with the nuclear genome. Due to their effect on traits such as metabolism and growth rates, cyto-nuclear interactions are affecting variation predicted to explain the evolution of behavioural types or personalities (i.e. behavioural variation that is consistent within individuals, but differs among individuals). However, while cyto-nuclear interactions have significant potential to explain variation in behaviours, this line of research remains poorly explored. We used nine cyto-nuclear integression lines, where three cytoplasmic genomes were introgressed into three nuclear backgrounds, to disentangle genetic effects on both life-history traits and behavioural variation in the seed beetle (Callosobruchus maculatus). We show that life-span, but also activity of individuals in behavioural assays are influenced by the interaction of nuclear and cytoplasmic genes. Variation in activity level is consistent among individual beetles, suggesting that intergenomic interactions can also explain variation in animal personality. These results advance our understanding of the functionality of mitochondrial genes and their non-neutrality, and highlight the importance of cyto-nuclear interactions in explaining variation in behaviour and personality.

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EVOLUTIONARY DIVERSIFICATION AT MULTIPLE LEVELS OF VARIATION IN DROSOPHILA WING SHAPE

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Studying integration and modularity is essential to understand the evolution of shape because the coherence of recognizable parts of most organisms is dependent on their developmental origin and structure. Drosophila wing morphology has been used extensively as an important model trait in evolutionary biology, since its genetics and development are well known. The aim of the present study is to address questions related to the evolution and developmental process of the wing shape architecture. Specifically, the questions are: 1- Are the Drosophila wings a single integrated morphological unit consistently in all the species groups across the whole genus? 2- Does morphological integration evolve across the genus? 3- Is there any evolutionary integration in the Drosophila wing? To study these morphological changes we used geometric morphometrics as a tool for analyzing shape variation and its covariation between the anterior and posterior (A/P) compartments of the wing and to compare the different covariance matrices between species. Moreover, we also used comparative methods for mapping the shape data onto the phylogeny. The analyses show, on the one hand, strong differences between the species of this genus. Indeed, the modularity test for the A/P wing compartments showed that they are not independent in all the species studied, confirming previous results for *D. melanogaster* at large scale. On the other hand, mapping the variation of covariance matrices onto the phylogeny shows the pattern of evolution of integration in the genus. This study has helped to answer questions related to the evolutionary process of the Drosophila wing shape and to show that the A/P wing compartments are not separate modules of variation. We have also found a clear evolutionary integration into all the different traits of wing shape. We can conclude that the wing shape shows strong internal covariation, and also that the integration process has evolved in the genus.

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EFFECT OF MALE REPRODUCTIVE PROTEINS ON FEMALE FITNESS IN SEED BEETLES

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In organisms with internal fertilization, males transfer seminal fluids to females during copulation. Specific proteins secreted by the male accessory glands (Acps) have profound impact in female reproductive physiology and behavior, affecting fecundity and female remating rate. Thus, these proteins may mediate postcopulatory sexual selection and may generate sexual conflict. Such selection can lead to their rapid evolution, with consequences for speciation. Despite their importance, Acps have been identified and their function elucidated in few cases and our understanding of the genetic variation in Acps production and the selection upon their encoding loci is still incomplete. The main goal of this study is to elucidate the level of genetic variation for Acps across populations in a seed beetle species, Callosobruchus maculatus, and to identify the Acps that mediate the effects upon female fitness through comparative quantitative proteomics studies. By using 7 different populations, we performed an experiment to estimate genetic variation in the Acps that regulate offspring production and female longevity. To obtain the Acp-proteome profile from each population we performed two-dimensional gel electrophoresis (2D-PAGE) and we generated the proteomic fingerprint needed for quantitative comparative analyses. Preliminary results showed that males from different populations have differentially affected female fecundity and longevity. Our results suggest the presence of genetic variation across populations for the effects of the Acps upon the studied female traits. 2D-proteomics comparative analyses showed differences in the Acp-proteome profile across populations, revealing a potential correlation between changes in the proteome and the female fitness. This finding is essential to identify the Acps that mediate the effects upon female fitness. This novel information will offer new insights into the evolution of Acps.

D23SY31PS1245

BEHAVIOURAL CONSISTENCY IN RANA TEMPORARIA TADPOLES: STRONG PERSONALITIES, LACK OF SYNDROMES AND LINK TO LIFE-HISTORY

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Recent interest in evolutionary behavioural ecology has turned towards understanding the causes and consequences of behavioural consistency, manifesting either as animal personality (i.e. consistency in a single behaviour) or behavioural syndromes (i.e. consistency across two or more behaviours). Further, behavioural consistency has been linked to life-history strategies currently, possibly forming integrative pace-of-life syndromes (POLS). However, empirical tests of this theory are extremely scarce. Here, we tested, using agile frog (Rana dalmatina) tadpoles as model, if POLS could be detected in the larval stage of amphibians based on a common garden experiment targeting two ontogenetic stages and measuring three behavioural traits. Activity was consistent in the early stage and was negatively related to age at metamorphosis. In the later stage, activity, exploration and risk-taking was all consistent, but their relationship with life-history depended on whether the studied individuals were tested in the earlier stage or not. Already tested individuals showed no POLS, but both activity (negatively) and risk-taking (positively) was related to age at metamorphosis in their previously untested conspecifics. We only detected one behavioural syndrome: activity and risk-taking was strongly negatively correlated in the previously tested (i.e. disturbed) group. Our results provide evidence for (i) animal personality in tadpoles, (ii) an activity – metamorphosis-timing POLS and (iii) an unexpectedly strong effect of moderate disturbance related to standard behavioural testing on later behavioural consistency.

D23SY31PS1307

HORMONAL EFFECTS ON THE VOCAL COMMUNICATION AND ANTI-PREDATOR BEHAVIOUR OF COOPERATIVE BREEDING MEERKATS (SURICATA SURICATTA)

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Animal vocalisations, like human voices, are characterised by individual differences in rate of production and acoustic structure. This variation can be partly explained by the social and environmental contexts that individuals experience, but also by their internal state. Thus, vocalisations can potentially be sources and vehicles of information concerning the producer's age, size, rank, sex and endocrine status. Individual differences in androgens (e.g. testosterone) and glucocorticoids (e.g. cortisol) have been repeatedly associated with competitive ability, social rank, vigilance behaviour and reproductive success. In social species, hormone levels have additionally been associated with variation in calling behaviour, including calling rates and acoustic parameters. Meerkats (Suricata suricatta) are small cooperative breeding mammals that live in groups of up to 50 individuals in Southern Africa. Meerkats have a highly developed vocal communication system, which they use to coordinate their spatial organisation, social interactions and anti-predator behaviour. Thus, meerkats are an ideal study system in which to explore links between behaviour, vocalisations and hormones in animal societies, using both natural observations and manipulations. Experimental elevation of glucocorticoids significantly increased the production rate of close calls in males but not in females. This may potentially reflect a greater role of subordinate males than females in the maintenance of group cohesion, where close calls seem to play a major role, in periods of greater predation risk. In addition, reduction of circulating testosterone levels in subordinate males tended to increase calling rate. Overall, our results show how androgens and glucocorticoids can affect vocal communication and behaviour in meerkats with potentially significant consequences to group cohesion and coordination of anti-predator behaviour in this species.

D23SY31PS1350

ANTHROPOGENIC DISTURBANCES ON THE DEVELOPMENT AND PHYSIOLOGY OF MARINE SPECIES

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Natural and human-induced disturbances across aquatic and terrestrial habitats affect a broad range of species and communities causing declines in both biodiversity and population abundance. Whilst habitat disturbances are known to affect species' distributions, phenology and physiology, less is known about the effects on development and stress physiology, especially for marine species. We investigated how a range of disturbances, mainly anthropogenic, affected the development and stress physiology of different tropical marine species. We will describe how two different habitat disturbances, the 1998 global bleaching event that reduced live coral cover and chronic boat noise, increased levels of the teleost stress hormone, cortisol in the skunk anemonefish and three-spot damselfish respectively. We will also describe how shark- and ray-feeding dive sites interact with insulin and gonadal steroid hormones of the black-tipped reef shark. Finally, we will describe how chronic boat noise slows sea hare embryonic development and reduces veliger hatching success. We will discuss how these varying habitat disturbances could impact reproduction and survival, highlighting their consequences for population dynamics.

D23SY31PS1385

A GENERAL MECHANISM FOR CONDITIONAL EXPRESSION OF SEXUALLY-SELECTED TRAITS

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Exaggerated sexually-selected traits are widespread across nature, with the "classic" examples including the trains of peacocks and antlers of cervids. These traits have provided the focus of many models and much debate concerning how they have evolved and how they are maintained. It is clear that these traits are reliable signals of the bearer's condition, but the underlying physiological mechanism required to achieve such exaggerated and highly condition dependent growth has remained, until recently, unclear. Handicap models have been proposed but lack consistent experimental support. Using the horned rhinoceros beetle, Trypoxylus dichotomus, we demonstrate that the insulin-like signaling (ILS) pathway can provide a robust and reliable physiological mechanism for generating an informative signal of individual condition. The ILS pathway is a highly conserved pathway that can regulate growth in response to nutritional level, stress and infection. Exaggerated and highly condition dependent growth in beetle horns is achieved by heightened sensitivity of the developing horn primorida to the ILS pathway, when compared to other traits such as wings and genitalia. This result, combined with evidence from a wide range of taxa (e.g. crustaceans, cervids and fish), as well as the highly conserved nature of the ILS pathway, raises the exciting possibility that parallel processes underlie the evolution of trait exaggeration in a multitude of sexually-selected signal traits across the animal kingdom.

D23SY31PS1423

ON THE CONDITION DEPENDENCE OF MALE GENITAL SPINES IN THE SEED BEETLE C. MACULATUS

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The rapid divergent evolution of genital morphology is largely attributable to postmating sexual selection through cryptic female choice. Yet, our understanding of the evolutionary mechanisms of cryptic female choice for male genital traits is very incomplete. In the seed beetle Callosobruchus maculatus previous research has shown that male reproductive fitness increases with genital spine length. Here, we investigate the effects of artificial selection for male genital spine length on female fitness. We first show that, remarkably, females from lines in which males were artificially selected for longer genital spines exhibited comparatively greater fitness (life-time reproductive success). One explanation for this is that male genital spines in *C. maculatus* are condition dependent. That is, selecting for longer male genital spines may indirectly select for male genetic quality, and differences in female fitness across these selection lines may be due to shared genetic architecture between the sexes. We test the hypothesis that male genital spines in C. maculatus are condition dependent by rearing genetically independent families (derived from the same base population as the selection lines) through a series of substrates that vary in nutritional value. Differences in genital spine size across developmental substrates indicate condition dependence of the trait. Variation in the degree of condition dependence across genetic families indicates standing genetic variation for condition dependence (G x E), which could account for the fact that artificial selection on male genitalia generates a correlated response in female fitness. We discuss our findings in terms of the origin and maintenance of traits selected via cryptic female choice.

D23SY31PS1438

REPEATED LAKE-STREAM DIVERGENCE IN STICKLEBACK LIFE HISTORY WITHIN A CENTRAL EUROPEAN LAKE BASIN

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Life history divergence between populations inhabiting ecologically distinct habitats might be a potent source of reproductive isolation, but has received little attention in the context of speciation. We test for life history divergence between threespine stickleback inhabiting Lake Constance (Central Europe) and multiple tributary streams. Otolith analysis show that lake fish generally reproduce at two years of age, while their conspecifics in all streams have shifted to a primarily annual life cycle. This divergence is paralleled by a striking and consistent reduction in body size and fecundity in stream fish relative to lake fish. Stomach content analysis suggests that life history divergence might reflect a genetic or plastic response to pelagic versus benthic foraging modes in the lake and the streams. Microsatellite and mitochondrial markers further reveal that life history shifts in the different streams have occurred independently following the colonization by Lake Constance stickleback, and indicate the presence of strong barriers to gene flow across at least some of the lake-stream habitat transitions. Given that body size is known to strongly influence stickleback mating behavior, these barriers might well be related to life history divergence. Current work is using high-resolution genome-wide marker data to explore genomic signatures of selection across the habitat transitions.

D23SY31PS1461

BRAIN SIZE AND PERSONALITY: ARTIFICIAL SELECTION REVEALS A POSITIVE GENETIC CORRELATION BETWEEN BRAIN SIZE AND PROACTIVITY IN THE GUPPY

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Animal personalities range from shy, cautious, easily stressed individuals ('reactive' personality types) to bold, innovative individuals that are prone to risk-taking and routine formation ('proactive' personality types). Though personality differences are expected to have important fitness consequences, their underlying mechanisms remain enigmatic. In this study, we investigated the relationship between individual differences in personality and variation in brain size. We performed three standard personality assays using populations of guppies (Poecilia reticulata) under artificial selection for large and small relative brain size, which also have known differences in cognitive ability. First, large-brained guppies were faster to habituate and more exploratory when tested in an open field; large-brained females were also bolder. Second, large-brained animals excreted less cortisol in a stressful confinement test. Finally, large-brained individuals were slower to adopt a novel food source, a result which is likely due to stronger routine formation rather than lack of innovation. Overall, our findings indicate that large-brained animals have a more proactive personality type, a characteristic which may affect important aspects of their ecology such as dispersal and niche exploration.

Symposium

32. Climate Change and Evolution

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3, 4

Organisers: Ary Hoffmann and Mauro Santos

Invited speakers: Ary Hoffmann

Description:

An increasing number of empirical studies and theoretical analyses consider the likelihood of populations evolving in response to the direct and indirect effects arising from rapid climate change. In this symposium we aim to provide an update of the recent literature. We also focus on studies that have investigated ways to increase evolutionary resilience in species particularly by altering patterns of gene flow within landscapes and by enhancing genetic variation within populations.

D20SY32IT18:33R2

CLIMATE CHANGE ADAPTATION: GENETIC AND GENOMIC APPROACHES IN DROSOPHILA

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It is being increasingly appreciated that rapid evolutionary changes can help species counter the negative effects of global warming, and also that they can allow species to exploit favourable conditions provided in a warming world. There are now several well documented cases of rapid genetic change in response to natural and experimental warming in animals and plants. The genetic and genomic basis of these changes can be understood through comparisons of populations. Moreover, the genes and genomic regions identified in these comparisons can be further investigated through functional analyses on model organisms and along environmental gradients. Because related species often differ in their evolutionary potential, there is also an opportunity to investigate the genomic basis of limits to climate change adaptation, particularly as more sequenced genomes become available. I illustrate the opportunities provided through this framework by considering recent research on Drosophila. However I also highlight limitations of these approaches for predicting the dynamics of adaptive shifts in populations. Ideally genetic and genomic approaches need to be combined with quantitative studies of selection in populations.

D20SY32RT10:30R2

POTENTIAL AND CONSTRAINS OF ADAPTATION TO OCEAN ACIDIFICATION IN THE GLOBALLY IMPORTANT MARINE PHYTOPLANKTON SPECIES EMILIANIA HUXLEYI

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The biological impacts of climate change and the associated acidification of the surface ocean are intensely studied, with likely consequences for biodiversity and ecosystem functioning emerging as a consensus. However, evolutionary responses have only recently been considered. Marine microbes with short generation times and large population size are good candidates for testing the evolutionary potential to respond to ocean acidification. We used the globally important marine phytoplankton species *Emiliania huxleyi* for a 1000 generation selection experiment in elevated CO2 and found that adaptive evolution to ocean acidification is possible and likely fast enough to act on time scales relevant to climate change. Replicate populations selected under high CO2 consistently revealed higher fitness than control populations under ocean acidification conditions. To investigate whether such phenotypic convergence involves the same or different mutations, we exposed adapted populations to a novel environment where pleiotropic effects can serve as a proxy for divergent genetic bases of previous adaptation to high CO2. We identified divergent functional genetic bases in replicate high CO2 adapted but not in control populations. This indicates that many evolutionary trajectories to high CO2 adaptation are possible in *E. huxleyi* but also suggests that pleiotropy may constrain adaptation of natural E. huxleyi populations to ocean acidification. Our results highlight the urgent need to consider evolutionary processes when assessing the responses of marine microbes to future ocean conditions.

D20SY32RT10:54R2

EVOLUTION ON THE MOVE: ADAPTATION TO USE A WIDESPREAD HOST ASSOCIATED WITH RESPONSES TO CLIMATE CHANGE IN THE UK BUTTERFLY ARICIA AGESTIS

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A species' geographical ranges typically consists of many populations differently adapted to local ecological conditions, such as the availability of particular hosts or prey. Specialization on localized resources could prevent species from colonizing new sites where the same resources are not available, limiting their capacity to track climatic changes. Consistent with this, we observe that rapid evolution of host preference as well as morphology related to increased dispersal has been associated with recent range expansion of the Brown Argus butterfly, *Aricia agestis*. While butterflies in long-established parts of their range exhibit local adaptation, usually showing increased preference for laying their eggs on the locally most abundant host plants, butterflies in recently-colonized areas show a consistent preference for a host plant species (Geranium molle) that is geographically widespread in the region of expansion, despite being locally rare. Reciprocal transplant experiments support these data, and show that recent colonists have lost local adaptations present in the established part of the range. Our data suggest that future anthropogenic warming can be expected to bring about a major restructuring of patterns of local adaptation, and may limit the potential for popuations to continue to evolve in the future.

32. Climate Change and Evolution

D20SY32RT11:18R2

ENVIRONMENTAL TOLERANCES, LATITUDINAL GRADIENTS AND THE POTENTIAL FOR SPECIATION IN MAMMALS AND BIRDS

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Janzen (1967) proposed that the broader temperature tolerances of temperate species should facilitate dispersal across the cold peaks of mountain barriers and therefore should promote latitudinal differences in the potential for speciation. This elegant hypothesis has since been extended to other environmental barriers to dispersal and suggested as a possible driver of latitudinal diversity gradients. Nevertheless, a key prediction –that gene flow across mountain passes, or other environmental gradients, is more likely in species with broader environmental tolerances– has never been tested in a large-scale comparative analysis that controls for the effects of phylogeny and other relevant factors. Here we test these ideas through phylogenetically-informed analyses of the potential for speciation in terrestrial mammals (3136 species) and birds (6694 species). We show that although greater tolerance to rainfall unpredictability reduces significantly the potential for speciation (as measured by the number of subspecies per species), greater tolerance to temperature variability has the opposite effect, even among species whose ranges are dissected by mountains. In addition, we show that the net potential for speciation is higher in temperate than tropical mammals, and that it does not vary significantly with latitude in birds. We discuss the implications of these findings for our understanding of the effects of climate change on global patterns of diversity.

D20SY32RT11:42R2

THE PUZZLE OF CNIDARIAN ADAPTATION: AN INTEGRATIVE APPROACH

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The ongoing climate change raises the question of the adaptability of species to heterogeneous and changing environment. This question is particularly important for long-lived and sessile species such as corals which may display low dispersal abilities. Studying adaptation and phenotypic plasticity in such species will help us understanding how they may produce adequate phenotypes allowing populations persistence or resilience. It is more and more clear today that studying these questions in these models requires considering all the partners of the coral considered as a complex meta-organism where several evolutionary units must be taken into account. Here we review the empirical evidences for diverse and interactive adaptive mechanisms playing in corals in the context of climate change. We report data generated at the community, population and individual levels and demonstrating how ecological conditions and life history of individuals influence adaptability. We also stress the putative role of acclimatization and epigenetic mechanisms in the response to stress and data concerning interindividual variations suggesting the potential for genetic adaptation. We discuss the relative importance, time scales and interactions of these processes for the persistence of coral populations. Considering these different data and ensuing hypothesis, we propose here an experimental framework allowing to disentangle the role of these different processes and of the different partners of the coral holobiont in adaptive ability of corals confronted to global changes.

32. Climate Change and Evolution

D20SY32RT14:00R2

A GENETIC AND ECOLOGICAL DISSECTION OF THE RECENT RANGE EXPANSION OF THE EUROPEAN WASP SPIDER ARGIOPE BRUENNICHI -CONSEQUENCE OF CLIMATE CHANGE?

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Rapid, poleward range expansions are observed for an increasing number of species and commonly attributed to global warming. The role of contemporary adaptation in facilitating such range shifts is often neglected and remains to be discussed. A typical example for a recent range shift is provided by the European wasp spider Argiope bruennichi. Originally limited to the Mediterranean region, the species has greatly expanded its range in the past century and can now be found as far north as Finland. Here we present a detailed phylogeographic analysis of the spider's range expansion, using mitochondrial DNA sequences, nuclear microsatellites and SNPs. We base our study on a dense, range wide sampling of more than 2000 contemporary samples and about 500 historical museum specimens from the time before the range expansion. The study is complemented by morphological data, thermal tolerance and -preference tests and a reciprocal transplant experiment of native and invasive European wasp spider populations. Using historical DNA, we can show that the spider's range expansion is accompanied by an increasing admixture of formerly isolated, genetic lineages. At the same time, morphological changes are evident in invasive spiders. The reciprocal transplant experiment indicates local adaptation of recently established populations to their colder overwintering conditions. Moreover, our experiments show that invasive spiders have lowered their temperature tolerance and -preference, indicating a shift of their temperature niche. Based on these results, we speculate that genetic admixture at the species range edge has recently enabled an adaptation of wasp spiders to cold Northern European climates. Currently, we are investigating the genomic signatures of this adaptation, using a whole genome sequencing approach along environmental gradients.

32. Climate Change and Evolution

D20SY32RT14:24R2

SEX IN MURKY WATERS: ANTHROPOGENIC DISTURBANCE OF SEXUAL SELECTION IN THE PIPEFISH MODEL SYSTEM

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Due to anthropogenic disturbance, both stable as well as naturally variable environments are becoming increasingly unpredictable. In order to persist, both opportunists and specialists need to respond to these changes. A variety of animal behaviours are thus affected by human induced environmental change, which in turn may alter species interactions, population dynamics and evolutionary processes. In marine ecosystems, overfishing, eutrophication, and ocean acidification is currently changing the aquatic environment at an unnatural speed. We use pipefish as our study organism, which is a family of highly specialized marine fish practicing male pregnancy. Through a series of controlled laboratory experiments we show that anthropogenic disturbance alter several aspects of reproduction, including the development of sexual ornaments, courtship behaviour, mate choice, mating propensity and reproductive success. We show that the way in which the environmental stressors alter reproductive behaviours may vary both between and within sexes, populations and species. Further, different environmental stressors, such as turbidity, hypoxia and altered pH levels, elicited different responses, sometimes in opposite directions. Thus our studies emphasises the complex way in which environmental change may alter reproductive behaviours. Given the increase in human disturbance on ecosystems, answering these questions is not only important to better understand how environmental fluctuations affect evolutionary processes, but also for conservation biology and studies of the resilience of organisms to anthropogenic induced environmental change.

D20SY32RT14:48R2

DO CLOSELY RELATED SPECIES USE THE SAME GENES TO ADAPT TO FLUCTUATING ENVIRONMENTAL CONDITIONS?

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For insects, seasonal fluctuating temperature conditions require the ability to acclimate to lower than optimal temperatures in order to remain active. Cold tolerance in insects has been shown to have a genetic basis in numerous groups; however it is not clear if the underlying genetic basis producing this phenotype is the same across species. We used the SOLiD platform to sequence the transcriptomes of two *Drosophila* species that diverged approximately 9 Myr ago, *D. montana* and *D. virilis*, from both cold acclimated and non-cold acclimated individuals. We established the genes which were differentially expressed between the treatments, allowing us to a) identify the genes and pathways involved in cold acclimately 150 genes were differentially expressed between the two species. Approximately 150 genes were differentially expressed between cold acclimated and non-cold acclimated *D. virilis* and *D. montana*. The biological processes associated with these genes were broadly similar in both species and included circadian clock regulation and metabolism. Despite being involved in broadly similar biological processes, few specific genes were found to be differentially expressed in both species. This suggests that the underlying genetic basis of cold acclimation in both species uses different genes, but similar pathways, to produce a similar phenotype.

D20SY32RT15:12R2

LOCAL ADAPTATION DRIVES LATITUDINAL FREQUENCY CLINES FOR CHROMOSOMAL INVERSIONS IN DROSOPHILA SUBOBSCURA

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Climate change is already impacting the distribution and behavior of many species. Nonetheless, population genetic changes in response to global warming have been observed only for a few species. This is the case of Drosophila subobscura, a native Palearctic species that harbors a rich chromosomal inversion polymorphism with "warm climate" inversions increasing in frequency worldwide. However, the selective process that maintains inversions in populations is not clear yet. The two main hypotheses differ in how the genetic content of inversions varies along a geographical gradient; the local adaptation hypothesis states that an inversion will have the same genetic content in all populations, while the coadaptation model suggests that in each population the genetic content will be different. In order to elucidate which model is more suitable for *D. subobscura*, we have analyzed ten genes -six of them are candidates for thermal adaptation- located on the longest and inversion richest chromosome O of the species. The most frequent chromosome arrangements in two Spanish populations along a latitudinal gradient were analyzed. No within-inversion genetic differences were detected among populations, which suggest that the gene content along the gradient is rather constant for the various gene arrangements. Although gene flux between different inversions (either by gene conversion or double crossover) was detected, significant genetic differentiation among inversions for all genes mapped within the inverted fragments of the chromosome was found.

D20SY32RT15:45R2

HOW RAPID CAN RAPID EVOLUTION BE? DIFFERENCES BETWEEN SEMI-ARID AND MEDITERRANEAN POPULATIONS OF BRACHYPODIUM DISTACHYON

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Models of future climate change predict a rapid decrease in precipitation and lower water availability in the East Mediterranean region. One consequence of this change will be a northward desertification process into the Mediterranean climate region, threatening the Mediterranean ecosystems. To survive this climate change, plants may adapt by a plastic response, or by an evolutionary-genetic response, which can be either genetic or epigenetic mechanisms. Understanding the rate and direction of evolutionary change of wild plants under climate change will enable better ecosystem management. Natural geo-climatic gradients provide a natural lab for studying the outcome of climate changes. Thus, the arid areas in Israel can serve as the predicted climate in the Mediterranean region following climate change. This study provides evidence for a rapid evolution of phenotypic traits following experimental rapid climate change. We tested different traits of the annual grass *Brachypodium distachyon* (Poaceae), related to the cereal crops, from Mediterranean and semi-arid sites in Israel. Seeds were collected from both sites after five and ten generations of experimental rain decrease (drought), and morphological and physiological traits were compared in common-garden conditions. We found differences in life-history and morphological traits after five generations only. We also found that while Mediterranean plants can undergo rapid evolution, semi-arid ones are pre-adapted. We argue that plants can adapt to future climate change and undergo a rapid evolution under strong environmental selection. These results are important not only in a conservation perspective but also in an agricultural perspective.

D20SY32RT16:09R2

DO RISING TEMPERATURES ACCELERATE THE EVOLUTIONARY ARMS RACE OF HOSTS AND PARASITES?

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Climate change is expected to impact host-parasite coevolution, but information on effects of climate variables on such systems is sparse. Here, three-spined sticklebacks (Gasterosteus aculeatus) infected with the tapeworm Schistocephalus solidus were experimentally exposed to different temperatures. Hosts and parasites were laboratory offspring of populations from Norway, Spain and Germany. Infections were done at 18°C and temperatures were changed to 13°, 24° or kept at 18°C. Whole transcriptome sequencing (RNAseq, Illumina) was performed with liver RNA from a subset (n=96) of control and infected sticklebacks maintained at 13° and 24°C. Temperature alone had significant effects on the gene expression. Exposure to the parasite boosted the temperature effects on genes expression. At 13°C S. solidus grew slower compared to 18° and 24°C, which might partially be explained by lower metabolic rates, but also by the higher activity of the stickleback's immune system at 13° compared to 18° and 24°C. This was supported by the gene ontology (GO) analysis of the RNAseq data, which revealed that immune genes of infected sticklebacks were more strongly differentially expressed in infected sticklebacks at 13° compared to 24°C. Parasite growth rates did not simply increase with temperature and were lower at 24° compared to 18°C. This suggests that immune activity of sticklebacks is optimal and represses parasite growth around 13°C, while S. solidus grows best at 18°C. Similar observations were made with different origins of sticklebacks and S. solidus, but Norwegian parasites were more virulent and Norwegian sticklebacks were more resistant than the other origins. In ectothermic systems, at first parasites benefit from rising temperatures, but over-exploitation of hosts at very high temperatures might become maladaptive also for the parasites. The present data suggest that rising temperatures indeed have the potential to accelerate the arms race of hosts and parasites.

D20SY32RT16:33R2

THE EVOLUTIONARY ADAPTIVE POTENTIAL OF DAPHNIA GALEATA IN TIME AND SPACE

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In the framework of global change research we aimed at estimating the impact of elevated water temperatures on an aquatic keystone species, the freshwater zooplankter *Daphnia galeata*. To quantify the evolutionary potential of the resident natural Daphnia populations, we performed experiments measuring variation in life-history traits under different temperatures. In a "resurrection ecology" approach, we sampled dormant egg banks of *Daphnia galeata* populations to obtain a representative sample of natural genetic diversity. We first compared locally adapted populations by sampling individuals from recent/superficial sediment layers, sampled along a latitudinal gradient from Norway to Italy. In a second experiment we used layers from different depths, comparing clones/populations from different time periods. After hatching and establishing clonal lineages in the laboratory, we performed common-garden experiments. D. galeata individuals from different time periods and latitudes were submitted to various water temperatures in flow-through-systems. In all experiments elevated water temperatures were shown to result in an increased somatic growth rate and accordingly an earlier age at maturity. In addition to the observed phenotypic plasticity, significant temperature x age interactions and temperature x origin interactions indicate thermal micro-evolution within last decades. With regard to these patterns of adaptation, we discuss the consequences of global warming on the fate of *Daphnia* populations and European freshwater biotas.

D20SY32RT16:57R2

MOLECULAR DATA AND ECOLOGICAL NICHE MODELING REVEAL THE EVOLUTIONARY HISTORY OF THE NEW CALEDONIAN RELICT AMBORELLA TRICHOPODA (AMBORELLACEAE)

<u>Valérie Poncet</u>¹, François Munoz¹, Jérôme Munzinger¹, Yohan Pillon¹, Céline Gomez¹, Marie Couderc¹, Christine Tranchant-Dubreuil¹, Serge Hamon¹, Alexandre DeKochko¹

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Amborella trichopoda Baill. is the sole living member of a lineage (Amborellaceae, Amborellales) that may be over 200 Million years old, long before the re-emergence of present New Caledonia. We investigated the biogeographic history of Amborella using a combined population genetics, species current distribution and palaeo distribution modelling approaches. We present here the first study carried out on Amborella trichopoda genetic and ecological variation in its natural distribution. We analyzed the pattern of genetic structure of 18 populations distributed throughout its geographical range. We found significant differentiation between any pair of locations, and four main welldifferentiated, geographically distinct genetic groups were inferred using the Bayesian clustering algorithm in Structure software. To assess and understand the spatial distribution of genetic diversity in relation with habitat suitability, we modeled the ecological niche of Amborella trichopoda using Maxent both in the present-day and during the Last Glacial Maximum (LGM) climatic conditions. The model based on current climatic and environmental conditions (Topography, Rainfall, Temperature, Pedology) fitted well the actual distribution of the species. The southern genetic group appears partly separated from the rest of the populations by an ecological barrier of ultramafic land. For the other groups, we found the cumulated habitat distances to be correlated with geographic distances, so that there was no barrier in suitable habitat that could explain the genetic differentiation of the groups. This suggests other biogeographic constraints, while the genetic differentiation within each of the groups clearly follows the isolation by distance model. Indeed, the spatial delineation of the largest genetic groups is consistent with the forests having been isolated for multiple glacial-interglacial cycles, as predicted by the LGM modeling.

D20SY32RT17:45R2

ENVIRONMENTAL VS. GEOGRAPHICAL MOLECULAR ADAPTATION IN ALPINE CONIFERS

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Alpine ecosystems are facing rapid human-induced environmental changes and more knowledge on the adaptive potential of keystone Alpine tree species is needed. This study addresses the question as to whether geographical isolation (IBD) or environmental-driven selection (IBA) is more important for adaptation, based on SNP genotyping of silver fir and larch natural populations in the Alps and Apennines. Genetic structure was tested for both geographical and environmental groups, using AMOVA. For each species, environmental groups were defined using climate variables selected from a multiple factor analysis. Complementary methods were applied to identify outliers based on these groups, and to test for IBD vs. IBA. AMOVA showed weak but significant genetic structure for both group types. Among the potential outliers detected, up to four loci were found for geographical groups and up to seven for environmental groups. A stronger effect of IBD than IBA was found in both species; nevertheless, once spatial effects were removed, elevation in fir and precipitation in larch were relevant factors explaining genetic structure. In Alpine trees, environmental-driven selection and geographical-isolation are both important for adaptation. This process is advantaged by large amount of seeds and strong selection at early stages of establishment.

D20SY32RT18:09R2

LONG-TERM LIFE-HISTORY RESPONSES TO CLIMATE CHANGE IN THE WILLOW WARBLER (PHYLLOSCOPUS TROCHILUS)

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Birds, and especially long-distance migrants, are excellent indicators of climate-induced phenological change. Advanced spring arrival is a typical response reported in a great number of species. Earlier arrival can lead to e.g. earlier egg-laying and thus have potentially great fitness consequences. Studies have suggested that certain aspects of avian life-history can predict a likely response to climate change. However, some responses, e.g. autumn migration, may vary extensively between species and specific life-history traits may even differ between populations and individuals. Autumn migration have been largely neglected in studies on breeding populations and most studies that investigate phenological shifts have focused on ringing data of individuals of unknown breeding population. To disentangle which life-history traits that need species-specific considerations in climate change contexts temporal data on breeding individuals is required. We present a long-term study on the autumn migratory behaviour of the Willow Warbler over a 25-year period in relation to climatic indices. Specifically, the change in autumn departure of a breeding population of the Island of Gotland in Sweden is considered. We found that the median appearance of juveniles in low moult status, i.e. individuals hatched on the island, have advanced by ca 9 days in 20 years. Migrating individuals have advanced their autumn median passage date by ca 5 days. Spring arrival and egg-laying dates on the island have similarly advanced since the 1990s, indicating a shift in breeding period. The advanced peak appearance of local juveniles suggests that this species, a long-distance migrant able to rear secondary broods, has not extended the breeding period at this location, e.g. in order to have secondary broods, but kept breeding period constant and shifted it forward in time.

D20SY32RT18:57R2

PATTERNS OF GENE FLOW AND FINE-SCALE GENETIC STRUCTURE IN PHENOLOGICAL-ISOLATED ALPINE POPULATIONS OF DWARF WILLOW (SALIX HERBACEA L.)

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Current threats to biodiversity, such as climate change, are thought to alter abiotic conditions and therefore intra-specific diversity within the highly heterogeneous alpine environments. Assessing the spatial organization and dynamics of genetic diversity present within species can help to predict the responses of organisms to environmental change. In this study, we evaluated whether snowmelt timing imposes restrictions for the gene flow between alpine microhabitats in the common long-lived alpineartic key species Salix herbacea L. 273 genets across 12 snowbed and ridge localities in the Swiss Alps were surveyed for phenological variation during 2 years and for genetic variation using 7 SSR markers. Differences in snow cover accounted for phenological differentiation among microhabitats of one month on average. Unexpectedly, gene flow appeared to occur freely among microhabitats, elevations, and adjacent mountains suggesting long-distance seed dispersal. Interestingly, ridge sites, which are supposed to resemble future alpine conditions, tended to exhibit lower levels of diversity than snowbeds. Also, patterns of effective population size (Ne) and migration (Nem) among microhabitats were strongly asymmetric, with ridges acting as sources and snowbeds as sinks. Since no recent genetic bottlenecks were detected in the studied populations, these asymmetric patterns are likely to reflect the current meta-population dynamics of the species, rather than ancient recolonization after the last glacial period. We discuss the consequences of these patterns of diversity and gene flow in the light of the extent of local adaptation and the ability of populations to respond to environmental change.

POSTERS

INSIGHTS INTO THE ECOLOGY AND BIOGEOGRAPHY OF AMBORELLA TRICHOPODA: INTRASPECIFIC GENETIC DIFFERENTIATION IN NEW CALEDONIA

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Amborella trichopoda Baill. is the sole living member of a lineage (Amborellaceae, Amborellales) that may be over 200 Million years old, long before the re-emergence of present New Caledonia. We investigated the biogeographic history of Amborella using a combined population genetics, species current distribution and palaeo distribution modelling approaches. We present here the first study carried out on Amborella trichopoda genetic and ecological variation in its natural distribution. We analyzed the pattern of genetic structure of 18 populations distributed throughout its geographical range. We found significant differentiation between any pair of locations, and four main welldifferentiated, geographically distinct genetic groups were inferred using the Bayesian clustering algorithm in Structure software. To assess and understand the spatial distribution of genetic diversity in relation with habitat suitability, we modeled the ecological niche of Amborella trichopoda using Maxent both in the present-day and during the Last Glacial Maximum (LGM) climatic conditions. The model based on current climatic and environmental conditions (Topography, Rainfall, Temperature, Pedology) fitted well the actual distribution of the species. The southern genetic group appears partly separated from the rest of the populations by an ecological barrier of ultramafic land. For the other groups, we found the cumulated habitat distances to be correlated with geographic distances, so that there was no barrier in suitable habitat that could explain the genetic differentiation of the groups. This suggests other biogeographic constraints, while the genetic differentiation within each of the groups clearly follows the isolation by distance model. Indeed, the spatial delineation of the largest genetic groups is consistent with the forests having been isolated for multiple glacial-interglacial cycles, as predicted by the LGM modeling.

GENETIC ARCHITECTURE AND PHENOTYPIC MISMATCH AT RANGE LIMITS IN COLOUR POLYMORPHIC DAMSELFLIES

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What does limit a species' distribution without any physical barriers? Theoretically, genetic load due to maladaptive gene flow from central populations or absence of genetic variation due to lack of gene flow are hypothesized to constrain adaptation to the novel environment in marginal populations, preventing range expansion. However, the links between gene flow and deviation of fitness-related traits from trait optima along environmental gradient remains largely unclear in nature. Here we examined population genetics and the degree of adaptation of fitness-related traits (body size and wing loading) along the latitudinal environmental gradient using two ecologically similar congeneric damselflies (Ischnura asiatica and I. senegalensis) in Japan which also differ in the absence (I. asiatica) or the presence (I. senegalensis) of female limited colour polymorphism. The former species has its distribution central to Japan and the latter is at its northern biogeographic boundary. At the overlapped geographic region in Japan, the monomorphic *I. asiatica* showed adaptive latitudinal cline in both body size and wing loading, whereas *I. senegalensis* showed maladaptive cline in these traits and the degree of maladaptiveness increased near the northern boundary of their distribution. Population genetic analyses found evidence for both asymmetric migration from the south (distribution core) and the loss of genetic diversity near the distribution boundary but only in *I. senegalensis*. Our study indicates the constraining effects of maladaptive gene flow and lack of genetic variation constrains adaptation at different spatial scales, and cooperatively prevents their range expansion in nature.

VANISHING CHROMOSOMAL INVERSION CLINES IN DROSOPHILA SUBOBSCURA FROM CHILE: IS BEHAVIORAL THERMOREGULATION TO BLAME?

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Chromosomal inversion clines paralleling the long standing ones in native Palaearctic populations of *Drosophila subobscura* evolved swiftly after this species invaded the Americas in the late 1970s and early 1980s. However, the new clines did not consistently continue to converge on the Old World baseline. Our recent survey of Chilean populations of *D. subobscura* shows that inversion clines have faded or even changed sign with latitude. Here we investigate the hypothesis that this fading of inversion clines might be due to the Bogert effect; namely, that flies' thermoregulatory behavior has eventually compensated for environmental variation in temperature, thus buffering selection on thermal-related traits. We show that latitude flies having a lower mean of thermal preference. Plastic responses in thermal preference also lessen latitudinal thermal variation because flies developed at colder temperatures prefer warmer microclimates. Our results are consistent with the idea that active behavioral thermoregulation might buffer environmental variation and reduce the potential impact of thermal selection on other traits as chromosomal arrangements.

HOST PLANT RANGE AND CLIMATE CHANGE: PREDICTIONS FOR A BUTTERFLY

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Although changes in distributions, phenology and species associations are relatively well documented responses to global warming, the potential interactions between these phenomena are less well understood. In this study we investigate the interactions between temperature, phenology and host plant use in the polyphagous butterfly *Polygonia c-album*. We found that the hierarchy in larval performance on three natural host plants was not modified by temperature, with growth rates above a minimum, allowing the host plant use range to be maintained. However, a potential shift from univoltinism to bivoltinism, because of a longer temporal window for reproduction, would modify host plant use, most likely due to differences in phenological responses of the three host plants. Hence, host range evolution in response to warming would in this species be highly contingent on whether the population undergoes a predicted shift from one to two generations. We conclude that warming can have significant effects on species associations, but that phenological responses among the species involved can lead to complex outcomes.

THERMAL ADAPTATION GENES: INVERSIONS, GENE FLUX AND SELECTION

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The patterns of genome organization can be influenced by recombination, which is an important evolutionary force. In general, presence of recombination is advantageous, as demonstrated by the evolution of sex in most of the complex organisms. However, recombination can be disadvantageous in some situations, breaking some adaptive combinations of genes. In the present study, we have focused on the study of six thermal candidate genes (*Pif1A*, *Abi*, *Sqd*, *Yrt*, *Atpα* and *Fmr1*) located across the O chromosome of *D. subobscura*, in relation to three adaptive arrangements (O3+4+1, O3+4+7 and O3+4) collected at Barcelona (Eastern Mediterranean) and Mt. Parnes, Greece (Eastern Mediterranean). Significant genetic differentiation using FST values was found between genes located within inversions (O1 and O7), which is consistent with reduced recombination between inversions. More strikingly, the significant genetic differentiation encountered for the $Atp\alpha$ gene, which is located outside these inversions, is due to fixed nonsynonymous changes between arrangement, and signs of positive selection were detected for the O3+4+7 arrangement. Selective sweeps associated to O1 and O7 inversions were dated in 0.04 and 0.06 Myr, respectively. The similarity of the $ATP\alpha$ protein sequences of O3+4 with that of D. madeirensis may reflect historic events. However, similarity at the protein level between O3+4+1 and O3+4+7 arrangements for the $Atp\alpha$ gene could be the consequence of independent substitutions arisen in the two lineages due to adaptation to similar environmental conditions.

FACING A WARMING WORLD: TEMPERATURE ADAPTATION AND COMPETITION WITH AN INVASIVE SPECIES

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Climate change is one of the main drivers of environmental change, with profound effects on ecology and evolution. Species differ in their tolerance limits, and in their abilities to disperse and to respond to a changing environment. This may lead to mismatches in species interactions or change competitive outcomes. Furthermore, a warming climate can aid non-native species to establish and become invasive, with potentially serious effects on communities and ecosystems. We studied the competitive outcome between the native snail Theodoxus fluviatilis and the thermo-tolerant exotic snail *Potamopyrgus antipodarum* under ambient and increased temperature conditions in a field experiment. The experiment was conducted in the Biotest Basin at the Swedish Baltic Sea coast, which receives warmed up cooling water from the Forsmark nuclear reactor plants. As a result, water temperatures have been increased by 4 to 10°C for more than 30 years. We were particularly interested in whether native snails from populations affected by cooling water discharge were better able to compete with the thermo-tolerant invader. The native *Theodoxus* showed counter-gradient variation, with cold-origin snails growing faster than warm-origin snails in both ambient and warm temperatures. Thermo-tolerant Potamopyrgus affected Theodoxus growth negatively, and the warm-origin snails were not better at competing with it. *Potamopyrgus* had very high population growth rates, especially in warm conditions, suggesting that this species may become invasive in the Baltic Sea when temperature increases. The combined pressure of temperature increase and competition may have negative consequences for the native snail species.

FLUCTUATING TEMPERATURE LEADS TO EVOLUTION OF THERMAL GENERALISM AND PRE-ADAPTATION TO NOVEL ENVIRONMENTS

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Climate change scenarios do not only expect elevated temperatures but also increased temperature fluctuations. Environmental fluctuations are suggested to select for low levels of plasticity in fitness that is also hypothesized to increase organisms' ability to invade novel environments and affect virulence of pathogens. We tested these hypotheses and show that across a range of temperatures, opportunistic bacterial pathogen Serratia marcescens that evolved in fluctuating temperature (daily variation between 24 and 38 °C, mean 31 °C), outperforms strains that evolved in constant temperature (31° C) across all measured temperatures. Their better growth was also evident in novel environments with parasitic viruses and predatory protozoans. However, the strains from fluctuating environment were less virulent to Drosophila melanogaster host. Therefore, whilst supporting the hypothesis that evolution in fluctuating environments is paired with tolerance to several novel environments, our results show that adapting to fluctuating environments can also be costly in terms of reduced virulence. Together these results suggest that thermal fluctuations driven by the climate change could affect not only species thermal tolerance but also species' invasiveness and virulence.

THE FATE OF CHROMOSOMAL INVERSION POLYMORPHISM DURING ADAPTATION TO A NOVEL ENVIRONMENT

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Chromosomal inversions are widespread in Drosophila with strong evidence supporting an adaptive cause for the evolution and maintenance of inversion polymorphisms. One emblematic example of the adaptive role associated with inversions is the repeatable clinal variation in inversions frequencies in different continents as happens in Drosophila subobscura. More recently, it has been found that these inversion polymorphisms are shifting as a response to global warming. Both local adaptation and gene flow may be involved, the latter possibly overcoming historical constraints. Nevertheless, it is still unclear what forces are important in shaping the evolution of inversion polymorphisms. An approach to this issue is to study the evolutionary dynamics of chromosomal inversions of populations initially differentiated along a cline, during adaptation to a novel, common environment in the absence of gene flow. We use this strategy analyzing laboratory adaptation in Drosophila subobscura founded from contrasting European latitudes. Will natural selection in the new environment overcome the initial historical differences, promoting convergence of inversion frequencies? During the first 25 generations of adaptation to a common environment we found that the polymorphism of chromosomal inversions was gradually reduced in all populations. We also found persistent differentiation between populations from contrasting latitudes, though these differences reduced with increasing generations in the new environment. While genetic drift seems to play an important role in inversion frequency changes, we also found consistent increase in frequency of specific inversions (initially in low frequencies) across replicate populations, which suggests that selection also played a role. Altogether this study indicates that, in the absence of gene flow, inversion polymorphism evolves under a balance of selection and genetic drift with historical constraints also playing an important role.

VARIABLE ASSOCIATION BETWEEN INVERSIONS AND WING TRAITS IN DROSOPHILA SUBOBSCURA: ARE THERE CONSISTENT DIFFERENCES BETWEEN CONTINENTS?

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Clinal variation for wing size and shape as well as for chromosomal inversion frequencies has been found in Drosophila subobscura from Europe as well as North and South America. Particularly it has been reported that standard arrangements increase in frequency towards higher latitudes, where flies are also bigger. In the New World the rapid evolution of body size clines as a follow up of clinal evolution of inversions, suggested that the wing traits cline had been driven by the inversion polymorphism. Previous studies, in favor of this hypothesis, found an intra-population association between wing traits (size and shape) and standard chromosomal inversions. Nevertheless, it was also found that the association (for shape) or details of it (for size) had opposite signs in one European population (Adraga) and one South American population (Puerto Montt). This is probably due to a bottleneck effect following the colonization of the Americas. Despite this interesting finding it is still unknown if this is a generalized difference between continents. To tackle this question we here tested for the consistency of the association between wing traits and inversions in three populations along the cline of Europe as well as South America. Surprisingly we found no clear association between wing size and the number of standard inversions in either continent. On the other hand, we confirmed that the previously reported negative association between wing shape and standard dose of Puerto Montt spread with latitude through other South America populations. This is not in agreement with the positive sign of the cline for both wing shape and inversions. Overall and contrary to previous indications, this study suggests that the inversion and wing clines in D. subobscura have evolved independently from each other.

COMPLEX RESPONSE IN LIFE HISTORY TRAITS OF THE BULB MITES IN ELEVATED THERMAL CONDITIONS – AN EXPERIMENTAL EVOLUTION APPROACH

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Temperature is a key environmental factor affecting almost all the aspects of life histories in ectotherms. The theory predicts that organisms grow faster, reach smaller sizes and produce smaller offspring when temperature increases. In addition, temperature changes, through their effects on metabolism, may also influence the expression of alternative reproductive phenotypes (ARP). Although many studies investigated reaction norms of life history traits in relation to temperature change, little is known about how these reaction norms evolve. In our study we subjected the bulb mites to experimental evolution in two temperature treatments: control and elevated. After 18 generations we measured adult body size, eggs size and development time of both treatments at control as well as at elevated temperature. Thus, we were able to distinguish genetic changes (the effect of selection temperature) from environmental effects. The ARP expression was recorded in each generation. We found that mites developed faster and reached smaller sizes at increased temperature, but genetic effects of thermal adaptation were not always parallel to the observed reaction norms revealing quite complex patterns of life history traits response to temperature. Despite smaller body sizes females laid larger eggs at higher temperature. This effect was more pronounced in animals evolving at elevated temperature. Evolution at increased temperature affected also ARP expression with the proportion of armored fighters decreasing from generation to generation. We propose that this could be the consequence of temperature sensitiveness of cost to benefits ratio of expressing ARPs.

CLIMATE CHANGE, HOST-PARASITE INTERACTIONS AND SPECIES INVASIONS: AN EXPERIMENT INITIATED BY NATURE

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Biological invasions can be considered as an experiment performed by nature. They offer a unique way to study adaptation to new environments with all facets of new biotic and abiotic challenges like climate change and disease. On top of that, rising temperatures and risk of disease can interact and intensify selection pressure on invasive species to immunologically adapt to local parasites. The invasion of the Pacific oyster, Crassostrea gigas into the North Sea, is an ideal system to study the interactive effects of climate change and disease. Two independent invasions lead to the establishment of two genetically distinct populations, that differ in their selective history of disease outbreaks. While the Southern population is frequently subjected to natural selection induced by oyster summer mortality, with mortality rates exceeding 60%, Northern populations and hybrids in a secondary contact zone have been spared so far. Here, the interaction of high temperatures and bacteria of the genus *Vibrio* are believed to be the main causative agents of such mortalities. To test for differential selection gradients within the two invasion waves, we infected wild and artificially bred oysters with allopatric and sympatric Vibrio splendidus strains at prevailing and proposed future water temperatures. Based on mortality rates, immune response, and bacterial infection loads we observed that at high water temperatures oysters were severely impacted by Vibrio infection. However, while we consistently observed specific host immunological adaptation to sympatric parasites also at lower temperatures, direct advantages in terms of host fitness could only be detected at high temperatures indicating that the selective environment can unveil otherwise cryptic patterns of local adaptation.

ECOLOGICAL NICHE EVOLUTION, PHYLOGEOGRAPHY AND THERMAL ADAPTATION STUDIES IN DROSOPHILA AMERICANA SUGGEST IT IS A PLASTIC SPECIES

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The study of ecological niche evolution is fundamental to understand how the environment influences species distributions and the role it plays in adaptation to divergent environments and species formation. Drosophila americana is widely distributed in North America. It comprises two chromosomal forms distributed in a north-south cline (chromosomes X/4 fused and non-fused, respectively), which is maintained by selection. Here we present a study of the ecological niche evolution and thermal adaptation of *D. americana* and the two chromosomal forms with the aim of understanding how they respond to a variety of environmental conditions. For this, we have used Ecological Niche Modelling, phylogeographic analysis and performance (locomotor and developmental time) experiments. Temperature is the environmental factor that contributes most to the ecological niches, although the relevance of precipitation is also high in the model of the Southern populations. Thermal performance experiments show no difference in the locomotor activity across a temperature range of 15° to 38°C between flies from the north and the south of its distribution. Finally, we have modelled the past distribution of the species; its range during the last glacial maximum (LGM) was reduced to the southernmost of North America, while they had a similar distribution during the last interglacial (LIG) as currently. Analysis of the demographic history, nevertheless, detects no bottlenecks during the LGM. These results suggest that *D. americana* displays a plastic adaptation to the different temperatures of their distribution and do not support niche conservatism in Drosophila.

ADAPTIVE POTENTIAL IN A SNAIL POPULATION

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Adaptation to climate conditions is crucial for the existence and distribution of species and knowledge of adaptive potential will help us to predict how populations will be affected by climate change. Within-population genetic variation is the prior requirement for adaptation to changing environmental conditions, e.g. global warming. Here we examined if genetic variation in the responses to environmental change exists in a freshwater snail *Lymnaea stagnalis*. We exposed maternal family-lines originating from a Swiss snail population to thermal conditions similar to those during heat waves and assessed its effect on snail reproduction and immunocompetence. We found that high temperature increased snail reproduction and reduced immunocompetence. In addition we found family-level variation in snail reproduction and in all measured immune traits. However, we did not find family-level variation in response to a rise in temperature in any trait we measured indicated by non-significant family by temperature interactions. This suggests limited genetic potential to adapt to the predicted increase in the frequency of heat waves owing to climate change.

CO-EVOLUTION OVERRIDES CLIMATE CHANGE IN CUCKOO AND REED WARBLER ARMS-RACE

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The evolution of host organisms is influenced by their parasites and vice versa, and a co-evolutionary arms race often develops between the two. Evolution is also influenced by climate, but the interaction between co-evolution and climate has largely been overlooked. Here, we show that the co-evolutionary arms race between the parasitic common cuckoo (Cuculus canorus) and its host, the reed warbler (Acrocephalus scirpaceus), overrides climatic factors in affecting the egg sizes of both species. Cuckoo egg volume decreased during the 20th century while reed warbler egg volume in parasitized nests increased, resulting in cuckoo eggs that were closer in size to those of the reed warbler. Egg volumes of sympatric reed warblers in non-parasitized nests, however, remained stable during that period. Cuckoo body size indices remained constant or increased over the same period, whereas reed warbler body size remained stable. Temperature and precipitation were uncorrelated with egg volume of either species. We suggest that cuckoos evolved to lay smaller eggs, to reduce rejection probability. Furthermore, our results point to the possibility that cuckoos may actively selected host nests with the largest host eggs. We suggest that cuckoos recently gained the upper hand in their co-evolutionary arms race with reed warblers, overriding the effects of recent climatic changes.

THE ROLE OF SEXUAL SELECTION DURING ADAPTATION TO A NOVEL, STRESSFUL ENVIRONMENT

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The interplay between natural and sexual selection has been widely studied, and although Darwin invoked sexual selection to explain traits that were clearly not naturally selected, it is unclear whether sexual selection facilitates adaptation to novel environments or not. Here we examined whether sexual selection promotes adaptation to a novel environment using the powerful method of experimental evolution. Using a fully factorial design, we established replicate *Drosophila simulans* populations in both standard and novel temperature environments, both with and without sexual selection. We found that sexual selection did not promote adaptation to the novel environment and this does not appear to be due to intralocus conflict constraining the evolution of the sexes. These results therefore support the original view of sexual selection as a non-adaptive process. However after 30 generations of selection we did find significantly higher fecundity in females evolving under elevated sexual selection. We will discuss possible explanations for this finding.

CLIMATE AND TREMATODE-MOLLUSK RELATIONSHIP

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In trematode-mollusk relationships, climate warming may affect cercaria larvae production, virulence and competition between parasite species. In the present study we investigated the host-parasite relationship of the bucephalid trematodes Rhipidocotyle fennica and R. campanula which are transmitted from the unionid bivalve A. anatina to the second host, cyprinid fish Rutilus rutilus, via cercariae produced in the bivalve host. Results suggest that the annual period of cercarial production has become markedly longer in both parasite species from 1980's to 2012-during the period of a climatic increase in temperature and length of summer. As compared to R. fennica, the parasite R. campanula started to produce cercariae much earlier in summer, resulting in a longer ceracariae production period annually. Early start of larval production was made possible in *R. campanula* by having cercaria production machinery 'on standby' throughout the year while R. fennica become inactive during winter. In line with this, *R. campanula* occurred in higher prevalences than *R. fennica* in northern areas, opposite to the south. The shorter summers towards north eventually limit the cercariae production period of the parasites, but due to the earlier onset of cercarial emission *R. campanula* is probably able to inhabit higher latitudes than *R. fennica*. However, *R. campanula* appeared to be more virulent than R. fennica (in terms of host survival and reproduction) which may be a cost of R. campanula's ability to start cercariae production early seasonally. Impact of experimental temperature increase on annual cercarial production was positive in *R. fennica* but zero or negative in *R*. campanula. Thus, climate warming should benefit more *R*. fennica, and expand its range to the north. To conclude, climate change will presumably affect geographic distribution and abundance of *Rhipidocotyle* parasites, as well as modify the evolution of their virulence.

CONSTRATING SIGNATURES OF THERMAL STABILITY EVOLUTION IN RAY-FINNED FISH MITOCHONDRIAL DNA-ENCODED PROTEINS

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Ray-finned fishes (actinopterygii) have successfully colonized almost all aquatic habitats, exploiting a wide range of temperature, salinity, oxygen level and hydrostatic pressure conditions. Temperature in particular is a strong environmental constraint driving adaptation, and several recent studies have detected signals of selection on mitochondrial genome along latitudinal or altitudinal gradients as well as an interaction between mitochondrial and nuclear genotypes in determining metabolic performance. Numerous actinopterygii complete mitochondrial genomes are now available and crystallographic structures have been determined for most of the 13 mitochondrial DNA-encoded proteins. Concomitantly, studies of the structural differences between mesophilic and thermophilic (optimal growth temperature > 50°C) microorganisms have provided useful insights into the general evolutionary patterns leading to increased protein thermal stability. In this context, we used all currently available actinopterygii mitochondrial genomes in relation with an environmental database (FishBase) to test whether there was an association between the thermal stability of mtDNA-encoded proteins and the environmental temperature commonly inhabited by the species. Several predictors of thermal stability (local structural entropy, amino acid compositional bias, hydrophobicity) were used with and without prior partitioning of the protein sequences into transmembrane and exposed regions. Phylogenetically independent contrasts were calculated to test the relationship between stability and environmental temperature. Signatures of increased predicted thermal stability with increasing environmental temperature were detected for some but not all of the proteins, thus suggesting that environmental temperature constrains the evolution of mtDNA-encoded protein stability in ray-finned fishes but that this constraint is not homogeneous among proteins.

LOCAL ADAPTATION TO CLIMATE CHANGE: A SEMI-NATURAL STUDY OF THE DROSOPHILA SUBOBSCURA MODEL SYSTEM

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Anthropogenic activities are accelerating the rate at which our environment is changing. Such rapid change threatens biodiversity as organisms struggle to cope with stressful environments, for example, increased temperature. To understand how species may adapt to human-driven climate change, we are studying the adaptive genetic responses to natural temperature changes in wild populations of the fruit fly, *Drosophila subobscura*. This species has a rich history in associating environmental gradients with clinal chromosomal inversion polymorphisms and clinal variations in morphological and physiological traits. To examine genes involved in local thermal adaptation, we used established outdoor enclosures of flies along a temperature (latitudinal) gradient at six European sites (from Sweden to Spain), spanning over 20 degrees of latitude. Flies were sampled under three natural thermal profiles relevant to each locations to investigate differentially expressed genes and changes in expressed allele frequencies between populations by using RNA-seq. The two extreme populations were reciprocally transplanted to distinguish plastic genetic responses from locally adaptive ones. Here we report our preliminary results on the identification of genetic loci potentially involved in the process of local climate adaptation. Such loci may represent genomic hotspots, including within chromosomal inversions, of selection in response to climate change.

INTER- AND INTRAPOPULATION HYBRIDS OF DROSOPHILA SUBOBSCURA UNDER TEMPERATURE STRESS DIFFER IN FITNESS COMPONENTS

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Temperature is one of the most important variables that determines distribution and abundance of species. In combination with genomic stress, such as inbreeding and/or hybridization, temperature as environmental stress may act synergistically leading to significant decrease of fitness components and deterioration of population survival. The consequences of climate change and habitat fragmentation on genetic structure should therefore be studied in combination rather than separately. We sampled *Drosophila subobscura* populations from two ecologically different habitats. One is from urban part of Belgrade (Botanical garden) where seasonal and daily temperature fluctuations can be substantial and another from Sicevo gorge with slightly higher average temperature and smaller temperature fluctuations. Isofemale lines (IL) from each population were established, and withinline, intrapopulation and interpopulation crosses were made. Progeny from these crosses was reared at low (16°C) and high (22°C) temperatures and fitness components (egg-to-adult viability and developmental time) of all groups were recorded. The obtained results suggest the population specific response to thermal stress. The crosses within ILs showed lower fitness after exposure to temperature stress which supports the hypothesis that superiority of heterozygous individuals is probably due to the increased metabolic efficiency.

CHARACTERIZATION OF EXPRESSION QUANTITATIVE TRAIT LOCI (EQTL) ASSOCIATED WITH RESPONSE TO THERMAL STRESS IN THE THREESPINE STICKLEBACK, GASTEROSTEUS ACULEATUS

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Ongoing climate change will expose populations to altered thermal regimens, which are likely to include more frequent and higher temperature maxima. Whether and how a resident population can withstand or adapt to these new conditions will depend on the genetic architecture underlying responses to temperature changes. This includes both coding genes and the regulatory regions that govern expression of these genes. Recent advances enabling the quantification of transcription levels for a large number of genes and the genotyping of many thousands of genetic markers throughout the genome, together with novel statistical methods, are facilitating the identification of such regulatory regions via expression quantitative trait locus (eQTL) analysis. The threespine stickleback (Gasterosteus aculeatus) is an important model organism in evolutionary biology. The species occurs as resident populations in a wide range of habitats with different thermal profiles. We used sticklebacks derived from the Baltic coast of Finland to identify and localize eQTLs underlying changes in gene expression in response to thermal stress. Experimental subjects comprised 600 individuals in 30 sibhalfsib families, half of which were subject to a thermal challenge immediately prior to tissue collection. Liver mRNA expression was subsequently quantified using Agilent custom microarrays. Over 2000 genes were found to be differentially expressed between control and treatment groups. Genotyping of the families by sequencing on the Illumina platform, in combination with the existing G. aculeatus genome, was used to generate a linkage map that included over 10,000 SNPs. This enabled us to characterize and explore the regulatory networks underlying these changes in gene expression.

SEX-SPECIFIC TOLERANCE TO A WARMED-UP ENVIRONMENT DISTORTS POPULATION SEX RATIO IN A SALMONID

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Environmental changes can influence major demographic parameters by affecting individual behavior, growth, or survival. In some fishes, water temperature can even influence sex determination or may create sex-specific selection pressures. We tested both possibilities in a declining population of grayling (Thymallus thymallus, Salmonidae) that suffers from a marked shift in the operational sex ratio from on average 65% males in earlier years to around 85% males during the last 20 years. We raised embryos and larvae at various temperatures and found no effect of temperature on sex determination within ecologically relevant temperatures. In parallel, we raised about 15,000 grayling under benign and comparatively cool conditions through their first summer, marked and released them into the wild, and caught some of them back 4 and 5 years later at their natural spawning place. Our treatment significantly reduced the sex ratio skew. We conclude that female juveniles of our study population are more susceptible than males to some major stressors of a warmed-up environment. Such sex-specific susceptibilities can distort operational sex ratios and contribute to population declines.

PALEODISTRIBUTION OF ALTITUDINAL ACTINOTE (LEPIDOPTERA: NYMPHALIDAE) SPECIES IN BRAZILIAN ATLANTIC RAINFOREST

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During the Pleistocene tropical forests passed through expansions and contraction cycles what has been explored for lowland Brazilian Atlantic Rainforest. On the other hand, little has been done on the consequences of climates cycle in the altitudinal environments of the Atlantic Forest. The aim of this work was simulate the effects of Pleistocene climate changes in altitudinal environments of Atlantic rainforest to generate hypothesis for further phylogeographic studies. We collected data on the distribution of *Actinote conspicua* and *Actinote dalmeidai*, two Nymphalidae (Insect: Lepidoptera) species of Atlantic Forest endemic of high altitudes along the south of Brazil. We used Maxent to model the actual distribution of this species using 20 variables from Worldclim Database, in 30 arcseconds resolution. The same database provided the data to model the species distribution at 21kya (the Last Glacial Maximum) and 120-140kya (the Last Inter-Glacial period). In Maxent we used 5 replicates with 10 "random", response curves and applied the Jackknife test for all variable estimates. The results showed that, compared to the current distribution, both species occupied a wider range at 21kya and a narrower one at 120-140kya. Next step will be a phylogeographic study to contribute to the understanding of the effects of Pleistocene climate fluctuations in high altitude species.

TRANSCRIPTOME RESPONSES TO THERMAL STRESS IN TWO IBERIAN SQUALIUS (CYPRINIDAE) SPECIES LIVING IN DISTINCT RIVER REGIMES

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Environmental conditions shape the distribution of organisms in nature. Particularly, freshwater fish are quite sensitive when exposed to variations in environmental conditions, since they often cannot escape to more suitable locations. Temperature is a keynote environmental factor that poses substantial challenges to ectotherms' survival and therefore species must adapt or otherwise perish. Fish from the Iberian Peninsula are exposed to seasonal and daily variations in water temperature. Particularly, species inhabiting the southern Iberian regions are exposed to harsher conditions, with higher temperatures during the dry season. Aiming to discover and characterize a large set of genes involved in thermal stress, two congeneric species, *Squalius carolitertii* and *Squalius torgalensis*, were studied as representatives of the northern and southern regions, respectively. Both species were exposed to two treatments, a control (18°C) and a stress (30°C) condition, and transcriptome sequencing of three organs was performed for both species. By comparing libraries of RNA-seq in the two conditions, in each species, genes differently expressed were identified. The study of these genes is important to understand the potential of fish to endure the extreme events driven by climate changes in their native rivers.

CLIMATE WARMING: WHO SUFFERS MORE IN A HOST-PARASITE

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Climate change can alter species distributions, interactions and community compositions. Further, it can accelerate evolutionary processes. In host-parasite interactions it is difficult to predict whether the parasite or the host benefits most from global warming, because milder winter periods or higher nighttime temperatures can cause changes in development or transmission rates and may support the host/parasite. This study focuses on parasite virulence and prevalence in the context of stressful temperature conditions. We examined different strains of the host species Paramecium tetraurelia, a common freshwater ciliate, infected with the obligate intracellular bacterium Caedibacter taeniospiralis. This parasite is located in the cytoplasm of its host and vertically transmitted. An experimental approach was used to investigate if parasite virulence, defined as host's fitness reduction caused by the parasite, is increasing under acute stress temperature conditions. Fitness of infected and uninfected P. tetraurelia was measured and the fitness-reduction/virulence was calculated at five different temperatures including a host stress temperature. The virulence caused by C. taeniospiralis was rising with higher temperature, but parasite prevalence was reduced at the host stress temperature. In an evolutionary context, the parasite could go extinct under acute stress conditions and the host would become cured of infection. Therefore, it was also tested how the parasite evolves under stress and optimum temperatures in long-term. The parasites that evolved under stress conditions seem to reduce costs to the host while having a higher prevalence compared to parasites that evolved under optimal conditions. Acute and intense stress might harm the parasite and seem to cause its extinction, but the parasite appears to be able to evolve and adapt to high-temperature stress conditions as caused by climate change.

GENETIC CONFLICT BETWEEN THE SEXES AND RATES OF ADAPTATION TO CLIMATE WARMING

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Intralocus sexual conflict (IASC) occurs when selection favors sex-specific optima in traits that have a shared genetic basis in males and females. This form of genetic conflict plays a key role in evolution and is likely to cause a gender load on population fitness. However, even though theory predicts that IASC may hamper adaptation under environmental change, we know very little about its potential role in affecting evolutionary rates under climate warming. We quantified the intersexual genetic correlation for fitness itself at a benign (29°C) and a stressful (36°C) temperature in the seed beetle Callosobruchus maculatus, by measuring offspring production of males and females from isofemale lines. In the studied population, there is a substantial gender load on fitness at the benign temperature, signified by a negative genetic correlation between male and female fitness. However, the correlation changed sign and became positive at stressful temperature conditions, suggesting that novel conditions may reduce genetic conflict between the sexes and increase rates of adaptation to climate warming. We estimate intersexual genetic correlations for, and sex-specific selection on, key traits likely to be involved in responses to climate warming (metabolic rate, lifespan, water loss, locomotor activity and body size). These data allow testing of two mechanisms that each predicts reduced conflict at stressful temperatures: 1) a release of cryptic genetic variation that has similar effects on fitness in both sexes and increases the opportunity for selection, and 2) a change and alignment of sex-specific selection differentials under stress.

THE EUROPEAN BROWN CRAB (CANCER PAGURUS) AS A MODEL TO STUDY LOCAL ADAPTATION IN HIGHLY DISTRIBUTIONAL MARINE DECAPODS

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In the past 50 years, surface temperature in the North Sea has risen in the range of 1.5-2.5°C and climate projections predict a further increase. Its impact on marine organisms is broad and their survival will depend on their resilience to cope with these changing conditions. In the brown crab (Cancer pagurus) we study how strongly stress tolerance towards temperature changes and associated pathogen stress is determined by phenotypic plasticity or limited by genetically based local adaptation. We analyze local populations originating from habitats along a latitudinal gradient from Southern Europe to the Sub-arctic. We link expression patterns from controlled experimental conditions on temperature with 454 ultra deep sequencing on transcriptomes to build a temperature-treatment specific EST library. Differently expressed genes together with genetic variations in ESTs in contrast to neutral microsattelite markers are used to differentiate between neutral genetic differences between populations and genetic adaptation. Our results give new insights in the source of local adaptation of a highly distributional marine species. In detail we analyze whether local populations can adapt to changed climate conditions, how genetic diversity influences their adaptation and whether future climate predictions may lead to distributional shifts for marine species such as Cancer pagurus.

D20SY32PS1448

A HOTSPOT ON FIRE: PREDICTED IMPACT OF CLIMATE CHANGE ON GENETIC DIVERSITY AND DISEASE TOLERANCE IN THE ENDANGERED APENNINE YELLOW-BELLIED TOAD BOMBINA PACHYPUS

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Climate change, emerging infectious diseases and levels of population genetic diversity can 'conspire' in yielding the decline and extinction of populations and species. We modeled species distribution and carried out a climate change vulnerability assessment for the endangered Bombina pachypus, an Italian endemic toad recently hypothesized to be faced with such 'conspiracy'. For this species, southern Italy is the hotspot of genetic diversity and of disease tolerance to the chytrid pathogen Batrachochytrium *dendrobatidis*. This area showed the highest bioclimatic suitability for *B. pachypus* based on data for the years 1951-2000. Forecasted species distribution to 2080, under various models and emission scenarios, suggests that this area will become progressively less suitable, or even unsuitable for B. pachypus. Thus, loosing most of its evolutionary potential and historical resources to cope with chytrid outbreaks, B. pachypus will face a high extinction risk in the near future. On the other hand, in the northern regions, where populations are almost genetically invariable and demographically inconsistent, habitat suitability is predicted to increase in the near future. We suggest that captive breeding programs aimed at supporting translocation of genetic diversity from southern into northern populations (i.e. genetic rescue), are the only conservation actions with non-null probability to counteract this negative trend, and to attempt avoiding the human-driven extinction of this threatened amphibian species.

Symposium

33. Population Ecology

20 August



Program

Tuesday 20 August

Session(s): 1, 2, 3, 4

Organisers: Ricardo Beldade and Michael Morrissey

Invited speakers: Jean Clobert and Michael Morrissey

Description:

This symposium will include evolutionary perspectives on population ecology themes such as dispersal and life cycles in populations and meta-populations. The symposium will focus on theoretical and empirical developments in dispersal and life-cycle research welcoming synthetic approaches that span across taxa within these themes.

33. Population Ecology

D20SY33IT10:30R5

DISPERSAL: CAUSES, MECHANISMS AND POPULATION CONSEQUENCES

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Dispersal is a three steps process. The social and non social environments can act either similarly or dissimilarly at all these stages. We will review the actual knowledge on dispersal ecology and evolution and try to sketch priorities for future research. We will also try to examine how dispersal might interact with global changes and in turn influence the dynamic of fragmented populations.

D20SY33IT14:00R5

SELECTION AND EVOLUTION OF CAUSALLY-COVARYING TRAITS

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Causal effects of traits on one another and on fitness can be viewed both as the source of covariance among traits, and as the mechanism of natural selection. I will discuss the inference of evolutionary parameters (selection, genetics) conditional on a priori hypotheses of causal relations among characters, i.e., path diagrams. Inference of selection based on Wright's path rules yields selection coefficients that differ in their fundamental biological meaning from the selection coefficients that are commonly used in evolutionary quantitative genetics. Path-based coefficients analogous to selection gradients may most directly reflect interpretations about selection that biologists seek to make. I seek to formalize the meaning of path-based inference of selection by obtaining an equation to predict evolutionary trajectories based on such inferences. I discuss path-based inference of non-linear selection and approaches for empirical application of the quantitative genetic theory of causally covarying traits.

D20SY33RT11:18R5

DISPERSAL IN A WORLD WITH EDGES: HYPERMOBILITY, INERTIA, AND THE POPULATION COST OF EVOLUTION

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Dispersal is a fundamental characteristic of organisms -- it determines the spatial, genetic, and demographic structure of population and is necessary for the persistence of any species. However, from the perspective of the individual, dispersal often carries substantial costs that can come in the currencies of energetics, time, risk and opportunities. Often the dispersal strategy favored by individuals will not be the strategy that would be 'best' for the population (best in terms of maximizing size or distribution). This leads to one of two results 1) individuals disperse less than is best for the population ("inertia") or 2) individuals disperse more than is best for the population ("hypermobility"). Models of dispersal are often constructed to avoid edge effects, however, the real world is full of edges. A major risk of dispersal, the possibility of landing in unfavorable habitat, becomes exacerbated in habitats that are not only potentially fragmented but also have sharp boundaries (e.g., an archipelago). One might therefore predict inertia to be a problem in worlds of small size where most habitat is near an edge. Here we construct a model to address these questions. We find that small worlds, small patch carrying capacities, and high dispersal mortality all lead to inertia whereas large worlds, large patch carrying capacities, and low dispersal mortality lead to hypermobility. We also show that although heterogeneity in individual dispersal behavior can increase long-distance dispersal events, this is not guaranteed to be so. In a subset of parameter space heterogeneity actually decreases the probability of long-distance dispersal, in contrast to previous thinking.

D20SY33RT11:42R5

CICHLID EVOLUTION IN CRATER LAKES AND ECOLOGICAL OPPORTUNITY

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Ecological opportunity fuels the generation of biodiversity. When empty habitats are colonized ecological release favors species niche expansion and eventually the divergence of taxa via intraspecific resource competition and character release. The very young cichlid fish radiations from the Nicaraguan lakes in Central America are a powerful model for the study of very rapid diversification. Multiple recently formed crater lakes exist in this area, and all of them have been independently colonized by a subset of the fish fauna from the larger and older close by Nicaraguan Great Lakes. This setting is excellent to test the idea of ecological and character release following ecological opportunity. To this end we have studied the entire fish fauna of several crater lakes and characterized the ecological role of each of the species. We investigate the consequence of the absence of some of these species in the community for the remaining species, and the ability of the latter to expand their niche, release their morphological design and ultimately speciate.

D20SY33RT14:48R5

A FLUCTUATING ENVIRONMENT DRIVES COEXISTENCE IN FIVE NON-POLLINATING FIG WASPS

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The principle of competitive exclusion states that species competing for identical resources cannot coexist, but this appears paradoxical given myriad ecologically similar competitors. Theory shows coexistence is possible if a fluctuating environment changes the competitive dominance of species, but only when environmental variation leads to non-linear or non-additive population growth. Nonadditivity facilitates coexistence when competition and environment covary in their effects on growth such that competition is weaker in poor environments. The variation required to facilitate coexistence is tvpically interpreted as a physical aspect of environment. We investigate a system that might be considered constant under such interpretations, but show a hidden source of temporal variability relying on fluctuating dispersal distances between resource patches. We model populations that use resources in discrete, ephemeral, patches. Such ephemeral patch systems often support many competing species. We frame our model with respect to the highly diverse non-pollinating fig wasp communities that oviposit and develop within fig fruit. Using numerical and individual-based models, we show that temporal storage of larval wasps leads to long-term community coexistence under a wide range of biologically realistic parameter values when wasps face a trade-off between dispersal ability and fecundity. We empirically test whether or not such a trade-off exists in a community of five competitor species of non-pollinators associated with Ficus petiolaris. We find strong evidence of a negative correlation between species dispersal abilities and fecundities, which we present as an extreme case in which a single fluctuating environmental variable appears to mediate coexistence in a community of competitors. We suggest that fluctuating environmental conditions may drive coexistence more generally, especially among competitors regularly dispersing between ephemerally available habitat patches.

D20SY33RT15:12R5

QUANTIFYING THE DEMOGRAPHIC COST OF SELECTION IN A CHANGING ENVIRONMENT

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The lag load, or reduction in mean fitness caused by incomplete adaptive tracking of a moving phenotypic optimum, is a key quantity in models of evolution in changing environments. Despite its central theoretical importance, empirical studies quantifying the lag load and the factors affecting it are lacking. Here we explore these issues in a Dutch population of great tits Parus major, where warming springs have generated a mismatch between annual breeding time and the timing of an important food resource, providing an ideal opportunity to examine how the resulting directional selection for earlier breeding time impacts population mean fitness and related demographic parameters. First, we show that interannual variation in mismatch over almost four decades has surprisingly not affected population growth, despite it having led to intensified directional selection. We demonstrate an important mechanism contributing to this uncoupling, whereby fitness losses associated with mismatch are counteracted by fitness gains due to relaxed competition. Next, we parameterised a quantitative genetic model to predict the theoretical 'critical rate of environmental change', beyond which increased maladaptation leads to population extinction. Results imply that even 'mild' rates of climate change would be close to the critical rate. However, individual-based simulations that account for both evolutionary processes and density dependence revealed that the expected time to extinction, although highly uncertain, is on the order or centuries, rather than decades. These findings imply that microevolution would only rescue the population from mild rates of sustained climate change. On the other hand, they also illustrate that considerable maladaptation can be demographically tolerated in the short term through density dependence, without immediate population declines, effectively 'buying time' for microevolution to restore adaptation until the environment (climate) stabilises.

D20SY33RT15:45R5

REPRODUCTIVE BARRIERS BETWEEN HELIANTHUS PETIOLARIS ECOTYPES

<u>Katherine L Ostevik</u>¹, Rose L Andrew¹, Sarah P Otto¹, Loren H Rieseberg¹ ¹Botany, Faculty of Sciences of the University of British Columbia, Canada ostevik@biodiversity.ubc.ca

Helianthus petiolaris is a widespread sunflower species that usually occupies sandy soils. However, at Great Sand Dunes National Park in Colorado, populations of *H. petiolaris* survive in large and active sand dunes. We consider these dune populations to be an ecotype of *H. petiolaris* and a potential incipient species. Accordingly, there is significant phenotypic differentiation and low but significant genetic differentiation between the dune and non-dune ecotypes. For example, dune seeds are 3 times heavier than non-dune seeds. In order to better understand the extent of speciation in this system, we measured reproductive barriers between the two ecotypes. We found asymmetric immigrant inviability, in which dune seedlings emerge and survive better in both habitat types, some assortitive mating via pollen competition, and weak intrinsic post-zygotic isolation. Interestingly, in our pollen competition experiment some plants showed biased seed set while others did not suggesting that 'choosiness' is polymorphic within the ecotypes of *H. petiolaris* are consistent with those we would expect between populations at an early stage of ecological speciation. Finally, we plan to compare adaptation and speciation in this system with two pairs of sunflower sister taxa with similar habitat differences.

D20SY33RT16:09R5

HYBRID SONGBIRDS EMPLOY INTERMEDIATE ROUTES IN MIGRATORY DIVIDE

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Seasonal migration may play a significant role in speciation; many divergent populations breed adjacent to one another while using different routes to reach their wintering grounds (i.e., form migratory divides). Migratory orientation is largely genetically determined in these populations and often involves navigation around geographic barriers. Accordingly, hybrids have been predicted to employ intermediate routes that are inferior to those of parental forms. We provide the first direct test of this hypothesis here, by attaching light-level geolocators to birds in a narrow hybrid zone between two groups of Swainson's thrushes that form a migratory divide in western North America. Most of these birds employed intermediate routes, navigating over large geographic barriers. The remainder employed a mixed strategy, using the same route as one parental form on fall migration and the other on spring migration. Data from age ratios and cline analyses further suggests that hybrids survive at lower rates than parental forms and are selected against. Together, these results provide strong support for the migratory divide hypothesis and represent one of few established examples in which a behavioral trait reduces hybrid fitness, thereby promoting speciation.

D20SY33RT16:33R5

CO-EVOLUTIONARY BRANCHING OF DISPERSAL AND SOCIALITY IN STRUCTURED POPULATIONS

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Dispersal has antagonistic effects for the evolution of altruism, cooperation and social behavior. On one hand, dispersal of individuals from their native patches decreases relatedness between locally interacting individuals, and thus disfavors the evolution of social traits. On the other hand, dispersal reduces local competition among kin, thereby favoring cooperation. It is traditionally thought that these two antagonistic effects balance, and as a consequence, an intermediate level of dispersion and sociality evolves. However, recent numerical experiments have suggested that when social traits and dispersal evolve together, evolutionary branching may take place, leading to the stable coexistence of social and asocial individuals with different dispersal strategies. In order to clarify the effects of dispersal on the evolution of cooperations. By tracking changes in the phenotypic distribution in a population over time, we are able to explicitly predict the conditions that lead to evolutionary branching, and thus the coexistence of different dispersal and cooperating strategies. More generally, our method can be applied to study the emergence of highly differentiated life histories in structured populations.

D20SY33RT16:57R5

DISENTANGLING THE SOCIAL, PARENTAL AND GENETIC INFLUENCES ON NATAL DISPERSAL IN GREAT TITS

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Natal dispersal is a key process underpinning the structure and dynamics of populations. Individual variation in dispersal behaviour is substantial, but we know very little about the causes of individual variation in dispersal, and the extent to which this variation is influenced by social processes. Here, we integrate longitudinal data collected over five decades from a wild great tit (Parus major) population with two cross-fostering experiments to disentangle the influence of social processes, parental effects and genetics on dispersal behaviour. We show first that parental dispersal phenotype, whether they are themselves locally hatched or immigrants, has scale-independent effects on dispersal by offspring. Birds with immigrant parents dispersed further within patches and were more likely to disperse outside patches. Using an index of the composition of early social environments, with reference to the immigrant and locally hatched status of neighbours, we then show that dispersal is independent of the local social environment in which birds are raised. We used two large-scale cross-fostering experiments to demonstrate that parental effects on dispersal are primarily intrinsic to offspring, and therefore independent of parental behaviour. Finally, we show that parental dispersal phenotypes show similar fledging success but differing rates of local recruitment of offspring, implying that dispersal phenotypes will be genetically structured across landscapes. Our findings suggest that understanding the underpinning genetics of dispersal will be important for understanding the behaviour of populations in fragmented landscapes. Non-random dispersal of particular types of individuals suggests scope for the emergence of fine-scale population structure and has important consequences for interpretations of selection studies and dispersal theory.

D20SY33RT17:45R5

FITNESS RETURNS OF INFLORESCENCE ARCHITECTURE IN A WIND POLLINATED ANNUAL PLANT

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Male fitness in outcrossing wind-pollinated plants is indirectly influenced by the efficiency of pollen delivery. Many wind-pollinated plant species present adaptations for efficient pollen dispersal. A typical example is provided by Mercurialis annua, where male individuals disperse pollen from erect inflorescences held above the plant. In contrast, hermaphrodite individuals usually release their pollen from sessile axillary inflorescences (axillary hermaphrodites), but pollen production and the length of inflorescences on hermaphrodites in some populations resembles that found in males (pedunculate hermaphrodites). The ability of males to invade and establish in populations with hermaphrodites is expected to depend directly on their relative ability to disperse pollen successfully. Thus, enhanced pollen production and dispersal ability by pedunculate hermaphrodites might prevent male invasion. Here, we estimated the pollen production and siring success of males and hermaphrodites of M. annua that varied in their inflorescence architecture in experimental mating arrays. Males sired four times more offspring when growing in populations with axillary hermaphrodites than with pedunculate hermaphrodites. We use these results, and those from previous work, to model the fate of males and hermaphrodites with different inflorescence architectures. Our results have important implications for the maintenance of males with hermaphrodites in this species (androdioecy). They also raise questions about the genetic architecture and the geographic distribution of inflorescence morphology in M. annua.

D20SY33RT18:09R5

THE IMPLICATIONS OF REVERSE SEX-BIASED DISPERSAL FOR SPATIAL GENETIC STRUCTURE IN A CO-OPERATIVELY BREEDING BIRD

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Understanding the extent to which individuals differ systematically in dispersal propensity is crucial for quantifying patterns of gene flow in wild populations. For example, sex-biased dispersal can give rise to vastly different spatial patterns of relatedness between sexes, whereby the philopatric sex has a much higher density of relatives in the immediate neighbourhood. Such patterns can generate population genetic structure, whilst in co-operatively-breeding species such differences in local relatedness could potentially underpin observed asymmetries in levels of intergroup-conflict. We present long-term ecological data from a co-operatively breeding bird to show that whilst both sexes show restricted dispersal, males disperse significantly further than females from their natal groups. We quantify the capacity of this dispersal bias to drive sex-differences in fine-scale spatial genetic structure by using a pedigree of 600 individuals in concert with these ecological data to partition gene flow into that caused by both dispersal and extra-group matings, the latter having the potential to reinforce or disrupt emerging patterns of genetic structure. We discuss our results in the context of their implications for our understanding of the processes shaping patterns of inter-group conflict, and genetic differentiation over small spatial scales, in wild populations.

D20SY33RT18:33R5

LIFE-HISTORY OF THE GLANVILLE FRITILLARY BUTTERFLY IN FRAGMENTED VERSUS CONTINUOUS LANDSCAPES

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Habitat loss and fragmentation threaten the long-term viability of innumerable species of plants and animals. At the same time, habitat fragmentation may impose strong natural selection on populations and lead to evolution of life-histories with possible consequences for demographic dynamics. The Baltic populations of the Glanville fritillary butterfly (Melitaea cinxia) inhabit landscapes with highly fragmented habitat (networks of small dry meadows) as well as landscapes with extensive continuous habitat (calcareous alvar grasslands). Here, we report the results of common garden studies on butterflies originating from two highly fragmented landscapes in Finland and Sweden and from two continuous landscapes in Sweden and Estonia, conducted in a large outdoor population cage and in the laboratory. We investigated a comprehensive set of 63 life-history traits, including components of larval development, adult behavior and reproductive performance. Habitat fragmentation shows association with several life-history traits. Most notably, the growth rate of the post-diapause larvae and several measures of flight capacity were higher in butterflies from fragmented than continuous landscapes. These results support theoretical predictions about high rate of population turnover in fragmented habitats selecting for increased rate of dispersal. Females from continuous landscapes had shorter intervals between consecutive egg clutches and somewhat higher life-time egg production, but shorter longevity, than females from fragmented landscapes. These results are likely to reflect the constant opportunities for oviposition in females living in continuous habitats, and possibly also the more dispersive females from fragmented landscapes allocating more resources to dispersal capacity at the cost of egg maturation rate

D20SY33RT18:57R5

SMALL FISH, BIG FISH: MODELLING ECOLOGICAL CONSEQUENCES OF LIFE-HISTORY EVOLUTION IN HARVESTED STOCKS

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An increasing amount of evidence shows that fishing and climate change are changing fish lifehistories. Empirically determined rates of change are usually within the range of 0.5-3% per year and they are positively correlated with fishing intensity, suggesting that some of the change is due to fisheries-induced evolution. Many fish now mature at younger age and smaller size and this trend is expected to continue. We used the marine ecosystem model Atlantis to explore how a slow (<0.1% per year) but continuous decrease in size-at-age of five harvested SE Australian fish species may affect their demography, species interactions and recovery potential. We found that even small decreases in fish sizes are amplified by positive feedback loops in the ecosystem and can lead to major changes in their natural mortalities. For some species, e.g. tiger flathead, a 4% decrease in length-at-age over 50 years resulted in a 50% increase in predation mortality. However, the magnitude and direction in predation mortality changes differed among shrinking species. The impact that shrinking of five species had on many functional groups in the ecosystem (change in biomass and diets) was similar and comparable to the impact caused by the introduction of moderate fishing into an unfished system. Some of the shrinking species failed to recover to the previous biomass levels even after fishing was completely stopped. The natural mortality of these species remained high, suggesting that the ecosystem had moved into a new state. Our model shows that human induced reduction in fish size will alter predation regimes, changing species interactions and strength of natural selection. The outcomes of this interplay between natural and fisheries induced selection on harvested stocks will determine the final rate of phenotypic change and is currently being explored in the ecosystem model that allows for the dynamic evolution of most species.

POSTERS

A NEWLY DISCOVERED ROLE OF EVOLUTION IN PUBLISHED CONSUMER-RESOURCE DYNAMICS

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The consumer-resource relationship is central to population biology and evolutionary ecology. While the interaction is conceptually simple, the mechanisms behind the persistence of any given consumerresource relationship vary and can be extremely complex. The interaction is generic: a majority of species participate as either consumer or resource, diner or dinner. Traditionally when population biologists interpret results, evolution is not considered to affect the population dynamics because ecological and evolutionary dynamics are assumed to occur on different time scales. Yet recent research has demonstrated that evolutionary change can happen rapidly and on the same time scale as ecological processes affecting population dynamics qualitatively. In here we present a reanalysis of 21 consumer-resource data published over the last 80 years, approximately. Of the 21 published studies of consumer-resource dynamics we analyzed, evolution played a significant and substantial role in ten and a marginally significant role in another three, even though the original authors did not recognize its effect. In addition to a reanalysis of previously published data, we demonstrate via mathematical modeling how rapid prey evolution results in a transition from predator-prey dynamics with classical quarter-lag cycles, to anti-phase oscillations typical of eco-evolutionary dynamics and at the extreme, to cryptic cycling. We then show that this mechanism very likely explains dynamics observed in historical data, where possibility of rapid evolution was originally ignored. In general, our reanalysis suggests that evolutionary dynamics occurred in many of these, but the significance of qualitative features in the dynamics went unrecognized at the time. Furthermore, with more prey generations and larger population sizes the likelihood of evolutionary dynamics was higher.

AGING AND IMPERFECT ASYMMETRIC DIVISION IN BACTERIA

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The process of aging is assumed to result from accumulating damage over the lifespan, and such damage finally leads to death. Evolutionary theory predicts that dividing organisms such as bacteria flee from that process at the population level by asymmetric division, that is one cell (presumably the mother cell) takes over more of the damage and the other cell (daughter) is perfectly rejuvenated. To test these arguments and to see whether asymmetric division remains perfect throughout life we conducted experiments on a single cell microfluidic bacteria system to compare demographic parameters of daughter cells that came from young mothers with daughter cells coming from old mothers. Results show that daughters from young mothers have long lifespan and divide more often compared to daughters coming from old mothers. There seem to be no differences in cell growth. Our results suggest that asymmetric division is only perfect early in life of cells but not late in life. Our findings can explain the maintenance of the cell line by perfect rejuvenation of daughter cells produced early in life, and at the same time see substantial aging processes. The results therefore combine previously contrasting theories of aging and show how life-histories have to be considered across generations.

DO PREY RESPOND APPROPRIATELY TO EVOLUTIONARILY NOVEL PREDATORS?

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Deserts provide a model system for studying convergent evolution. Sand dunes in the Mojave (USA) and Negev (Israel) deserts are home to distinct yet convergent community of species with granivorous rodents and sidewinding vipers, red foxes and barn owls as predators. By virtue of heat sensing pits(vipers) and external cheek pouches (hetromyid rodents) the Mojave possess constraint breaking adaptations over the counterpart species in the Negev. In a semi-natural arena, we exposed Merriam's kangaroo rat (*Dipodomys merriami*) and desert pocket mice (*Chaetodipus penicillatus*) of the Mojave, and Allenby's gerbil (Gerbillus andersoni allenbyi) of the Negev to sidewinder rattlesnakes (Crotalus cereastes), a pit viper, and to the Saharan horned viper (*Cerastes cerastes*). We quantified the response of each rodent species to the snakes and in combination with owls. We monitored activity over all moon phases and asked whether the rodents responded differently to the novel, compared to the familiar, snake species. Specifically, we test the recognition of gerbils of a novel predator with heat sensing pits, and heteromyids' assessment of a snake with limited tools.
Analysis shows that the gerbiline rodents, at first, risk encounters with the novel viper before rapidly learning to avoid both vipers. Lengthy exposure allowed gerbils to adjust and optimize their balancing of food and safety. This caused a 25% increase in foraging for both species of vipers. Pocket mice reduced their foraging equally regardless of viper species. Interestingly Kangaroo rats found both species equally frightening following exposure they ceased to be aversive of the novel predator.

EVOLUTIONARILY CONVERGENT RODENTS DO NOT CONVERGE ON PREDATOR EVASION STRATEGY

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Merriam's kangaroo rat (*Dipodomys merriami*) and desert pocket mice (*Chaetodipus penicillatus*) of the Mojave Desert and Allenby's gerbil (Gerbillus andersoni allenbyi) of the Negev Desert are granivorous rodents inhabiting desert sand dunes. These dunes are home to the sidewinder rattlesnake (North America) and the horned viper (a sidewinding snake of the Middle East). The gerbils in the Negev evolved with the Saharan horned viper (Cerastes cerastes) and the Mojave Desert rodents evolved with the sidewinder rattlesnake (Crotalus cerasts). Both predators and prey species in the Mojave are convergent to the Negev counterparts: The snakes have heat sensing pits and the rodents have fur lined cheek pouches. We exposed the three rodent species to both snake species in a test system and measured the inherent knowledge they possess on the predators. The process was repeated after two months of exposure to both snakes to understand what knowledge was acquired when the predation risk was real and the selective pressures it puts on the population. We were interested in seeing how the species view the evolutionarily know and novel predators, and to assess whether the rodents learnt the strengths and weaknesses of the snakes in relation to each other. We found that each species employs a different strategy in response to the predators. Gerbils learnt that the pit vipers are dangerous by decreasing the amount of time they spend foraging in their presence by 80% and forage 25% less food in those patches. Desert pocket mice avoid snakes overall and decreased the time they spend in the presence of the known predator (the pit viper) by 90%. Lastly Kangaroo rats employed an "in your face" predator management strategy. They doubled their foraging time in the presence of the novel predator with an increase of 13% in foraged grain. This suggests that these species who are functionally convergent may not be behaviorally convergent.

THE EVOLUTION OF FITNESS ASSOCIATED DISPERSAL IN HOMOGENEOUS ENVIRONMENTS

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Dispersal is a major factor in ecological and evolutionary dynamics, but the evolution of dispersal strategies is still puzzling, in particular when the cost of dispersal is high. Empirical evidence show that the tendency to disperse varies among individuals in many organisms. Which individuals, therefore, should disperse and which should stay? We suggest that dispersal can be viewed as a means to genetically diversify offspring when individual fitness is low, even in a homogeneous and stable environment. Using stochastic simulations we show that throughout the parameter range selection is likely to favor plasticity in dispersal - where fit individuals are less likely to disperse (Fitness Associated Dispersal or FAD) - over uniform dispersal (where everyone disperse with the same probability). Moreover, FAD can evolve under a much wider parameter range than uniform dispersal, even when the cost of dispersal is high, and such dispersal strategy has significant long term effects. We thus predict that individuals of low quality would have an increased tendency for dispersal, even when dispersal brings significant costs.

SPATIAL GENETIC STRUCTURE AND REPRODUCTIVE SUCCESS OF A PARASITOID WASP IN A COMPETITIVE ENVIRONMENT

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In presence of a limiting resource, individuals can suffer from intraspecific exploitation competition. In the context of a parasitoid foraging for hosts, the effect of competition on fitness can be seen as a variation in reproductive success. The parasitoid wasp Hyposoter horticola is a specialist of the butterfly *Melitaea cinxia*. The wasp parasitizes the host larva just before it hatches from the eggshell. The host is therefore available for parasitism for a limited amount of time only (a couple of days). To cope with this time constraint, female wasps start to forage for host eggs before the eggs are ready to be parasitized. Hyposoter horticola is a good forager; females monitor several host clusters and visit them regularly until they are susceptible to parasitism. This behaviour increases the amount of time available for parasitism but, since the location of a host cluster is known by several wasps and each cluster can only be parasitized by a single wasp, it creates competition among the foraging females. In the Åland archipelago, Finland, the host butterfly lives as a metapopulation inhabiting a subset of 4 000 patches of suitable habitat. In order to conduct genetic analyses of the parasitoid population, we developed 14 microsatellite markers and genotyped 800 individuals collected from each of the host local populations during 2009. Performing clustering and relatedness analyses, we assessed the genetic structure of the population as well as the extent of the landscape area covered by females during foraging for hosts. We also analysed the effect of environment (host population locality, size and patch connectivity) and competition (local host and parasitoid density) on the proportion of females that reproduce and their reproductive success.

DENSITY-DEPENDENT DISPERSAL IN SABLE ISLAND HORSES: RATE AND DIRECTION VARIES WITH SEX AND AGE BUT ALSO SCALE AT WHICH DENSITY IS ASSESSED

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Density-dependent dispersal is known for most species; why it evolves is of fundamental interest to ecology and evolution. In this context, we have yet to fully appreciate how individuals of varying conditions might differently perceive quorum in their decision whether to disperse, which may be scale-dependent. Using an individual-based field study, we demonstrate how dispersal rate in an island population of feral horses (*Equus ferus caballus*, Sable Island, Canada, 2008–2010) is positively density-dependent for all age and sex categories with respect to local density (breeding adults within 8000 m of a horse's centroid location), but negatively density-dependent for males and positively density-dependent for females in relation to group (band) size. Dispersal was generally female-biased, with the exception of foals which moved with their mothers (no sex effect), and for yearlings and subadults when band sizes were smaller than average, in which case males dispersed at higher rates than females. Dispersal distance was positively related to local density. We conclude that dispersal rate can be both positively and negatively density-dependent for feral horses, contingent on the state of individuals and the scale at which quorum is assessed. Scale effects and interactions of density-dependent and sex- and age-biased dispersal may have both ecological and evolutionary consequences through effects on resource and mate competition.

GENETIC VARIATION IN THE EFFECT OF MONOAMINES ON FEMALE MATING RECEPTIVITY AND OVIPOSITION IN THE ADZUKI BEAN BEETLE (CALLOSOBRUCHUS CHINENSIS)

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Female mate choice after mating is a strong force in sexual selection and would lead to co-evolution of mating traits between the sexes. Some genotypes differ in the female mating rate and fecundity, and these differences are thought to be caused by the female's mate choice in response to the male ejaculate, including substances in male seminal fluids. Monoamines regulate animal physiology and behavior, including the post-mating behavior of females. In the present study, I compared the effects of monoamines on female mating receptivity and oviposition among four Callosobruchus chinensis strains (populations) (isC, akC02, mC, and the previously studied jC-S strain). Injection with octopamine or tyramine solutions significantly reduced female receptivity in the jC-S and mC strains, with low remating frequencies. However, no monoamines reduced female receptivity in isC, with a high remating frequency. Oviposition was activated by tyramine or serotonin in jC-S and by tyramine in mC. However, octopamine activated oviposition in isC and no monoamines did in akC02. Previous researches showed that jC-S and mC females had stronger responses than isC and akC02 females to male seminal substances that reduce receptivity or induce oviposition, suggesting that similarities exist between the effects of monoamines, especially tyramine, and the responses to male substances. These findings suggest relationships between the differences in female responses to male seminal substances among populations and the existence of one or more mutations in the biogenic biosynthetic or transmission pathways, which in turn would cause female mate choice to respond to male substances.

REGIONAL AND TEMPORAL VARIATION IN THE BIODIVERSITY OF DROSOPHILA-PARASITOID COMMUNITIES IN EASTERN NORTH AMERICA: IMPLICATIONS FOR COEVOLUTION

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Coevolution, resulting from ecological interactions among species in communities, is thought to be one of the most important processes organizing biodiversity on Earth. Since coevolution requires long-term interactions between species, ecological factors that alter these interactions may reduce the likelihood of coevolution. My research is focused on understanding how temporal and geographic variation in community composition alters species interactions and so influences the process of coevolution, using Drosophila host species and their parasitoid wasps as a model system. As the first step in the study, we are chronologically surveying the biodiversity of natural Drosophila-parasitoid communities along a North-South cline in eastern North America at three different time points during the breeding season (from May to October, depending on location) of 2012. We have found extensive variation in Drosophila host species diversity and the results indicate that their biodiversity displayed geographic and temporal variation among five sample sites. We are currently in the process of identifying the parasitoids wasps from each sample period. Combining data from seasonal and regional variation of host-parasitoid community structure will lay the foundation for future experiments to explore how different regional ecological interactions influence the coevolution of host and parasitoid species.

GENETIC DIVERSITY PATTERNS OF MACROBENTHIC POPULATIONS FROM LAGOONAL ECOSYSTEMS

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Coastal lagoons are dynamic ecosystems characterized by vigorously changing temporal and spatial conditions. They are an intriguing field for the study of genetic adaptations and community behavior. In these ecosystems, a small number of species is able to tolerate these conditions. Polychaetes are one of the most abundant taxa inhabiting lagoons and in many cases, they have been found to provide a good assessment of the benthic community showing comparable trends to those produced by the total macrobenthic fauna. The aim of the study is to compare polychaete species population patterns from Amvrakikos Gulf (W Greece) and Ria Formosa (S Portugal) inferred from mtDNA data. The current study attempts to correlate genetic diversity patterns to the patterns of abiotic data from each lagoonal system. The research leading to these results received funding from the European Community's Seventh Framework Programme (FP7/2007-2013) under grant agreement n° 227799- ASSEMBLE.

DISPERSAL, FLIGHT METABOLISM AND FLIGHT-INDUCED GENE EXPRESSION IN THE GLANVILLE FRITILLARY BUTTERFLY

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Flight is essential for reproduction and dispersal in butterflies and other flying insects. Studies on the Glanville fritillary butterfly have shown that the higher the rate of flight metabolism an individual can achieve, the higher is its capacity for dispersal in the field. However, thoracic muscles of flying insects exhibit the highest mass-specific rates of oxygen consumption known for any locomotor tissue, making insect flight an energetically demanding activity. Flight that is based on aerobic metabolism may become quickly limited by oxygen conductance to the flight muscles, leading to hypoxia. In this study, we investigate genome-wide gene expression induced by controlled active flight in the Glanville fritillary. We aim to identify genes and pathways that are up- or down-regulated in flight, and we study how allelic variation in annotated genes, gene expression, and flight metabolism phenotypes covary. We compare females and males, which exhibit significant differences in their flight metabolism and movement behaviors, reflecting different functions of flight in the two sexes. Second, we contrast individuals from two natural populations with very different genetic compositions, a large metapopulation in the Åland Islands and a small, completely isolated island population with low genetic variability, high genetic load as well as greatly reduced flight metabolic rate.

CHEMICAL ANTHROPOGENIC POLLUTION INTERFERES IN PREY RECOGNITION SYSTEMS

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Chemical detection of predator cues is crucial for aquatic prey, because it allows predator avoidance and activation of plastic antipredatory defenses. This is the case of many larval amphibians that respond to water-borne cues from potential predators by strongly reducing activity levels. However, as a consequence of increasing anthropogenic activity, a variety of harmful contaminants are dumped in freshwater ecosystems, where they can create interferences in the cue recognition system of tadpoles. Here we analyze the potential effects of two contaminants (i. e., humic acid and ammonium nitrate) on the ability of tadpoles of the western spadefoot toad (Pelobates cultripes) to recognize chemical cues from a common predator, nymphs of the dragonfly Anax imperator. We compared swimming activity of tadpoles in presence and absence of water-borne chemical cues from dragonflies, at different concentrations of humic acid and ammonium nitrate. Changes in tadpole activity associated to toxicity were non-significant. In contrast, the interaction between predator cues and presence of contaminants was significant: tadpoles effectively responded to predator cues in the absence of pollutants by reducing their swimming activity, but remained unresponsive to predator cues when either humic acid or ammonium nitrate were added, even at low concentrations. These interferences due to chemical anthropogenic pollution may pose a threat to the cue recognition systems of prey-predator interactions evolved in amphibian populations.

GENETIC DIVERSITY AND STRUCTURE OF OECEOCLADES MACULATA (LINDL.) LINDL. POPULATIONS

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Originally from Africa, the terrestrial orchid Oeceoclades maculata (Lindl.) Lindl is considered an invasive species with potential to colonize large areas. The species grows well in dry and wet environments as well as is present in disturbed and undisturbed areas. Although presenting a passive mechanism of self-fertilization, studies also point to the occurrence of cross-fertilization in Brazilian populations, indicating the existence of variation in the reproductive biology of the species throughout its geographic range. Considering the lack of information on the population genetics of this species, the aim of the study was to assess the genetic diversity and structure of Brazilian populations of O. maculata and to obtain more information on the reproductive system of the species. Five populations were considered for analysis [three from São Paulo state (SP), one from Mato Grosso (MT) and one from Paraná (PR)]. Population genetic analyses were obtained from eight ISSR primers and 153 specimens. The percentage of polymorphic loci (P) for each population was variable: Piracicaba/SP (6.25%), Ribeirão Preto/SP (1.79%), Maringá/PR (12.50%), Piracicaba1/SP (34.82%), Cáceres/MT (2.68%). The values of Shannon index (I) and Nei's genetic diversity (He) were low, varying from I =0.0023, *He* = 0.0010 (Ribeirão Preto/SP) to *I* = 0.0305, *He* = 0.0160 (Maringá/PR). A cluster analysis made with Jaccard similarity coefficient and UPGMA method separated the five populations into five independent groups. The analysis of molecular variance (AMOVA) showed that 90.4% of the variation occurs between populations, indicating high genetic structuring, with more genetic variation partitioned among populations than was detected within populations. A Bayesian model-based clustering analysis placed the individuals into nine clusters, showing evidence that the groups are strongly structured. These results corroborate with other autogamous and invasive species.

SIZE-ASSORTATIVE MATING IN THE ANT *MYRMICA RUGINODIS* AND ITS IMPLICATION FOR SPECIATION

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Size-assortative mating occurs widely in animals - increasing the frequency of desirable traits and enhancing fitness. In queens of the myrmecine ant Myrmica ruginodis, two distinct morphs exist, larger (macrogyne) and smaller (microgyne) queens. However, whether the occurrence of these morphs is maintained by size-assortative mating is unknown. Females are considered to choose their mating partner during nuptial flights, while males compete over females and large males usually regarded as higher quality will find a mating partner more likely. Nevertheless, whether a bi-modal size distribution exists in males at all, and whether small males participate in a nuptial flight has not been confirmed so far. To test whether assortative mating occurs in the two queen-morphs in M. ruginodis, individual males as well as males and females engaged in copula were collected during nuptial flights, and measured for size. Furthermore, queens of excavated nests and their offspring were analyzed to investigate if size characteristics are heritable and therefore fulfill preconditions for an ongoing speciation. We investigated whether size-assortative mating occurs for the mating couples, as well as if spatial and temporal isolation of macrogyne and microgyne morphs during the mating flights exist. This study provides insight into the composition of nuptial flights of Myrmica ruginodis and their mating behavior in connection to the queen size dimorphism and its implications on speciation in this ant.

INCREASED RISK OF KIN COMPETITION INDUCES KIN AVOIDANCE DURING SHOALING BEHAVIOR

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Inclusive fitness theory predicts that individuals can indirectly increase their fitness by kin-biased behavior. However, kin-bias can also increase the competition between kin, potentially eliminating the benefits. Thus, individuals are expected to plastically adjust kin-bias according to the risk and intensity of kin-competition which can be highly variable depending on environmental conditions. We investigate the impact of competition on kin-bias in a shoaling context using juveniles of the cichlid fish Pelvicachromis taeniatus. Kin-shoaling is a widespread adaptation to reduce the predation risk in prey organisms. In the present study, variation in competition was increased by limiting food availability in greatly size-variable juveniles that were given the choice between a shoal consisting of kin and a shoal consisting of non-kin. Considering that food limitation affects differently sized fish differentially due to the extent of energy expenditure, the level of competition. Shoaling behavior switched from kin-preference at low competition levels to kin-avoidance during high competition levels. We suggest that individuals may increase their inclusive fitness by avoiding competition with kin.

RAINSPLASH SPORE DISPERSAL IN COLLETOTRICHUM GLOEOSPORIOIDES INFECTING YAMS: DISPERSAL DISTANCE AND EFFECT OF GROUND CONTAMINATION

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Pathogens evolved a great diversity of dispersal mechanisms, from biological vectors to wind or rain, or even a combination of these. Rain splash is known from a variety of plant pathogenic fungi, and may be an efficient drive of local epidemics in crops, whereby infectious strains can quite rapidly propagate to genetically homogenous neighbouring plants. Splashing is nevertheless a very local dispersal process and spores taking the droplet ride are bound to a vicinity of a few decimeters at best. In this study, we assessed rainsplash dispersal of conidiospores of the Yam anthracnose disease agent, *Colletotrichum gloeosporioides*, in an experimental setting using a rain simulator, with emphasis on the impact of prior soil contamination (effect of re-splashing events in spore dispersal). Inoculum source from leaves allows for a dispersal up to 50cm, though with an exponential decrease in distance traveled from source, while ground driven contamination via re-splash is weak but contributes more and more to dispersal with increasing distance from initial source. Our results demonstrate that recurrent rains may contribute to a much greater dispersal distance than acknowledged from single rain release estimates, and may efficiently contribute to local field epidemics in tropical environments.

MALE-MALE AGGRESSION PEAKS AT INTERMEDIATE RELATEDNESS IN A SOCIAL SPIDER MITE

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Inclusive fitness theory predicts that when individuals live in groups or colonies, male-male aggression should peak at intermediate levels of average relatedness in the colony. Assuming that aggression is costly and directed towards nonrelatives and that competition for reproduction acts within the colony, the benefits of aggressive behaviour are maximized in colonies with a mix of related and unrelated competitors because aggression hurts unrelated individuals often, thereby favouring reproduction of related individuals. This prediction has been tested with specific bacterial strains in laboratory settings, but not with organisms in the field. Here, we study how male-male aggression varies with average relatedness in naturally occurring colonies of the social spider mite Stigmaeopsis miscanthi. This mite lives on Chinese silver grass, constructs colonies as woven nests on the undersurface of grass leaves, and lives within these nests in kin groups. This mite species shows parental care: males defend the offspring they fathered by counterattacking the offspring from predatory mites that intruded the nest. However, males also show aggression towards conspecific males. They kill rival males inside nests, and establish their own harem. Male-male aggression (quantified as the probability of lethal combat) is highly variable among populations, and it is negatively correlated with a proxy for average genetic relatedness in the colony - winter harshness. We sampled 25 populations across a wide geographic range between Taiwan and Japan, representing a gradient of high to low within-colony relatedness. For each population the weaponry of males was measured as the length of the first pair of legs, and malemale aggression was tested by placing pairs of non-sibling males together and scoring the frequency of male death over a given period. In support of theory, male-male aggression and weapon size strongly peak at intermediate average relatedness.

THE POPULATION ECOLOGICAL FUNDAMENT FOR LIFE HISTORY EVOLUTION

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Although rarely recognized, population ecology may be the most essential natural selection fundament for life history evolution. I illustrate how population growth and intra-specific density dependent competitive interactions unfold from self-replication in limited environments generating selection for the inevitable evolution of organized organisms similar to those on Earth. This include the evolution of exponentially increasing body masses, the evolution of body mass allometries, and evolutionary transitions from the negligible-sized asexual self-replicator, over large bodies organisms with sexual reproduction, to the fully evolved eusocial colony.

DISPERSAL AND EVOLUTION OF THE EUROPEAN EEL SHAPED BY VARIATIONS OF NORTH ATLANTIC DRIFT CURRENTS

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For open ocean broadcast spawners, environment-dependent dispersal is a crucial but critical life phase. The yearly recruitment fluctuations and cryptic adult population structure commonly observed in these species are assumed to reflect both the temporal and the spatial environmental conditions that early stages are exposed to. With a long-lasting ocean-dependent dispersal, the European eel (*Anguilla anguilla*) poses as perfect model organism to study the role of the environment in the evolutionary ecology of a broadcast spawner. To this end, we compared predictions made on dispersal, recruitment and population genetics by 45 years of high-resolution North Atlantic drift modeling with natural recruitment and population genetics at European coasts. We identify atmospherically-driven current variations in the Sargasso Sea as a key factor for a successful dispersal. We also demonstrate how unexpected findings of population structuring are consistent with contemporary recruitment levels, and with cryptic female philopatric behaviors within the Sargasso Sea. We suggest this strategy to be an evolutionary consequence of the unpredictable oceanic environment in the Sargasso Sea.

GENETIC STRUCTURE OF THE ENDANGERED AMERINDIAN YAM (DIOSCOREA TRIFIDA L.)

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Dioscorea trifida (Dioscoreaceae) is an endangered Amerindian yam species, whose origin, evolution and domestication are still unresolved issues. In order to assist in developing strategies for the conservation of D. trifida, we assessed the genetic diversity of 53 accessions of D. trifida from the states of São Paulo, Santa Catarina, Mato Grosso and Amazonas in Brazil based on eight SSR and 16 ISSR markers. The level of polymorphism among the accessions was high, 95% for SSR and 76% for ISSR. The SSR marker showed higher discrimination power among accessions compared to ISSR, with D parameter values of 0.79 and 0.44, respectively. Although SSR and ISSR markers led to dendrograms with different topologies, both separated the accessions into three main groups: I -Ubatuba-SP; II - Iguape-SP and Santa Catarina; and III - Mato Grosso. The two accessions from Amazonas varied their position according to the genomic region analyzed. Bayesian and principal coordinate analyses conducted with both molecular markers corroborated the classification into three main groups. Higher variation was found within groups in the AMOVA analysis for both markers (66.5% and 60.6% for ISSR and SSR, respectively), and higher Shannon diversity index was found for data from SSR marker. Significant correlations were found between genetic and geographic distances (r = 0.08; p = 0.0007 for SSR and r = 0.16; p = 0.0002 for ISSR). Therefore, results from both markers showed a high spatially structured genetic diversity in D. trifida accessions maintained by small traditional farmers in Brazil.

NATURE'S ECOLOGICAL RECORDERS: STABLE ISOTOPES IN MYCALESINE BUTTERFLIES

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Mycalesine butterflies have radiated dramatically in Africa, Madagascar and Asia to produce more than 250 extant species. The primary driving process of these radiations could have been the ability of mycalesine butterflies to invade the empty niches that arose as a result of the evolution of the C4 photosynthetic pathway and the world-wide expansion of C4 grasses in open, sunlit environments. Primary forest species of mycalesines are expected to be C3 specialists as the advantage of the C4 pathway declines in shaded forest understories where cool conditions improve the relative photosynthetic efficiency of C3 grass species. C4 grasses are characteristic of more open, sunlit habitats and where we expect mycalesines to be more opportunistic and generalist in their host plant choice or even to have become C4 specialists. Reliable host plant data is essential to be able to test this hypothesis but host plant records for grass-feeding mycalesines are very limited. However, over the last three decades stable isotope analyses have become an important part of the ecologist's toolbox. We used a community of three sympatric species of *Bicyclus* in Malawi, which were caught on a daily basis through two wet-dry seasonal cycles between January 1996 and December 1997, to explore whether $\delta 13C$ the $\delta 18O$ can be used to detect micro-climate conditions during the larval development of Mycalesine butterflies. Preliminary data shows that the feeding history of the larvae, with respect to C3 and C4 grasses, can be traced by analysing the relative amount of 13C in the leg tissue of adults. Indeed, both savannah species primarily develop on C4 grasses whereas the forest species mainly uses C3 grasses. Increased evaporation of water in the insect is reflected by enriched d18O values in adult tissue. Our data clearly show that the d18O values in butterfly leg tissue are enriched during the dry season. There are interspecific differences that can be explained by adult habitat preferences.

GENETIC DIVERSITY OF COCOYAM (XANTHOSOMA SPP.) IN BRAZIL, ASSESSED WITH ISSR MARKERS

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The native American cocoyam (Xanthosoma spp.) is considered an important food crop in many parts of the tropics. The usable parts of these species are the subterranean tuberous stems and, as a secondary use, the young leaves, similar to spinach. This study aimed at characterizing the genetic diversity of local varieties of cocoyam collected from several regions in Brazil, using Inter Simple Sequence Repeat (ISSR) markers. A total of 31 accessions were analyzed with five ISSR primers, with 12 accessions from UNESP collection and 19 accessions from the National Institute for Amazonian Research (INPA). Three accessions from INPA were classified as *Colocasia* spp. Binary data was used to construct a Jaccard similarity coefficient matrix and data were analyzed using the PAST software. The classification of the accessions into groups was performed by conducting both a cluster and a principal coordinate analysis. A total of 89 bands were observed, of which 99% were polymorphic. Cluster analysis showed high genetic divergence among the accessions, with the Jaccard coefficient ranging from 0.06 to 1.0. The accessions were divided into two groups, one of *Xanthosoma* sp. and the other of *Colocasia* sp., with a *Xanthosoma* sp. sample that may be incorrectly identified. Within the *Xanthosoma* group, 3 subgroups were found, with 2 outliers, one of which was *X. blandum*. Accessions of *X. robustum*, *X. sagittifolium* and *X. violaceum* were mixed in the 3 subgroups. Three duplicates were observed, one of them including two accessions classified as X. violaceum, which grouped together with a X. sagittifolium accession. Accessions from the UNESP collection were separated in both analyses from the INPA collection, with one exception. No geographic clustering was found among the accessions. Analyses with a larger number of markers are being conducted in order to confirm the results. Financial support: FAPESP and CNPq.

POPULATION GENETIC ANALYSES OF THE ENDANGERED BRAZILIAN ATLANTIC FOREST 'CATTLEYA COCCINEA' ORCHID COMPLEX

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The '*Cattleya coccinea*' orchid clade comprise a high altitude species complex found in the mountain ranges of South and Southeastern Brazil. The identification of species in this group is challenging due to the overlap of morphological characters. Our goalwas to conduct population genetics analyses to describestructure and patterns of diversity within and among populations and thus understand lineage diversification. A previous study with ISSR markers showed high structuring and low diversity within populations. This study was performed with populations from eight localities and six SSR markers, widening geographical and morphological sampling to the southern and westernmost limits of the clade distribution. Results confirmed populations as highly structured(G'ST= 0.221) but indicated moderate to high levels of within-population genetic diversity (He= 0.482-0.703), possibly due to co-dominant marker sensitivity. Bayesian clustering analysis revealed four genetically distinct groups. The same four groups were also obtained in the cluster analysis using Nei's (1978) genetic distance, corroborating the previous analysis entirely. Our data does not corroborate current species delimitation. Instead threeeco-geographical lineages emergeas putative species, respectively from (1) northeastern Brazilian Atlantic Forest (BAF); (2) southern BAF; and (3) a westernmost lineage found in drier habitats, mainly 'campos rupestres'. High population structure is concordant with limited gene flow promoted by reward hummingbird pollination, suggesting genetic drift as an important mechanism of speciation. Additional population sampling and complementary phylogeographical and morphological studies are in progress to understand the contribution of drift and natural selection to the diversification within this clade. Financial support: FAPESP.

UNMATEDNESS PROMOTES THE EVOLUTION OF HELPING MORE IN DIPLODIPLOIDS THAN IN HAPLODIPLOIDS

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One major question in evolutionary biology is how non-reproductive workers have evolved in eusocial organisms. The 'haplodiploid hypothesis' attempted to explain why eusociality seems to have evolved more times in haplodiploids than in non-clonal diplodiploids. That theory was proven inadequate by Trivers and Hare (1976) who showed, however, that haplodiploidy could promote the evolution of eusociality provided the existence of variation in sex ratio among different broods ('split sex ratios'). Gardner et al. (2012) reviewed the empirical progress in the understanding of the evolution of eusociality, and suggested that the potentially important causes of split sex ratios at the origin of helping are limited to queen virginity (some queens not mating and producing all-male broods) and queen replacement. After Gardner et al's correction on the role of queen replacement has been taken into account, only queen virginity seems to be a possible cause for split sex ratios at the origin of helping in haplodiploids. However, when comparing haplodiploid and diplodiploid systems, Gardner et al. neglect the fact that unmated diplodiploid females have zero reproductive success. The analysis we propose reveals that unmatedness promotes the evolution of helping more in diplodiploids than in haplodiploids. Considering altogether Gardner et al's results and our own, we conclude that the factors causing split sex ratios in haplodiploids do not make eusociality more likely to evolve in haplodiploids than in diplodiploids.

GENETIC DIVERSITY AND GENE FLOW OF INTRODUCED POPULATIONS OF DYCKIA DISTACHYA HASSLER, A BROMELIAD EXTREMELY ENDANGERED

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Specific of rheophytics environments, *Dyckia distachya* is a rare bromeliad. Due to anthropic pressures imposed on its natural habitat, remains only one known population in the wild. Aiming to avoid their extinction, populations were introduced in similar places to those of natural occurrence. The main objectives are the verification of the structuring of introduced populations, inference of gene flow and evaluation of the genetic diversity between generations. The natural population and 4 introduced populations were evaluated using SSR nuclear markers. Samples were collected from all reproductive individuals in each population, being considered 15 mother plants by population and 15 individuals per mother plant. Parameters of genetic diversity from mother plants were calculated, such as observed and expected heterozygosities in Hardy-Weinberg Equilibrium (HWE) and inbreeding coefficient. Also we calculated the outcrossing rate, crossing between related individuals, paternity correlation and effective number of pollen donors. The observed heterozygosity in introduced populations was greater than natural, ranging from 0.292 to 0.406, while in natural was 0.169. All populations analyzed deviated from HWE. The inbreeding coefficient ranged from 0.247 to 0.307 in introduced populations, while the natural was 0.087. The outcrossing rate (1.072) indicates that there is high cross pollination in the introduced population and low in natural (0.214). The outcrossing rate between related individuals in the introduced population (-0.005) indicates that there are few inbreeding crosses and the paternity correlation (0.141) that the crossings occur preferentially among unrelated individuals. The number of half sibs in the introduced population was high (0.92), probably due to the effective number of pollen donors (7). The monitoring of introduced populations with genetic parameters will be important to elucidate some aspects of the introduction of species in nature.

FACTORS AFFECTING JUVENILE MOVEMENT IN A HOUSE SPARROW METAPOPULATION – THE ROLE OF TIME OF BIRTH AND HABITAT CONFIGURATION

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Dispersal propensity may vary considerably between individuals in a population. Still, dispersal remains one of the least understood processes in ecology and evolution. In this study, we investigate individual variation in juvenile movement during the onset of the dispersal phase in four insular house sparrows *Passer domesticus* populations in northern Norway. The predictors were clutch size, hatch day, sex and morphology. The study islands differ in degree of patch isolation, configuration of suitable habitat, and characteristics of the matrix habitat. Thus, it was possible to assess the importance of such landscape features on movement. On one of the islands, movement rate was negatively related to hatch day. This pattern may be generated by age-dependent movement since early born individuals are older when the dispersal phase is initiated. However, we suggest that this relationship rather is a result of seasonal variation in the environment experienced by the nestlings, which affects their phenotype and propensity to disperse. Clutch size, sex, body size, mass and condition did not significantly explain variation in movement. On the other hand, movement rate seemed to decrease with increasing patch isolation, i.e. distance between patches and hostility of the matrix habitat. This suggests that habitat configuration is an important factor influencing movement and dispersal capacities of juvenile house sparrows.

SEASONAL MIGRATIONS AND SPACE USE OF NON HUNTED MOOSE POPULATION IN THE BIEBRZA MARSHES POLAND

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The Biebrza marshes are one of the most important refuges for moose in Poland, where since 2001 temporary moose-hunting moratorium is imposed. The Biebrza population numbers over 600 individuals and is genetically distinct from other populations in Poland. In 2011, we initiated a project on space use and foraging ecology of moose with the use of modern methods: GPS-tracking and DNA-based diet analysis. We immobilised 21 moose (11 females and 10 males) in January-March 2012, and fitted them with GPS-GSM collars (Ecotone, Poland). Collars collect 24 locations a day. Our preliminary results showed that in winter, moose ranges (MCP 100%) covered an average area of 12.5 km2 (range: 3.2 - 30.2 km2) and did not differ significantly between males and females. Mean daily movement distance was 1,68 km (range: 0.78 - 2.12 km). Collared animals utilised mainly pine forests. The majority of moose (67%) migrated a short distance (7.0 km on average; range; 3.3 to 18.8 km) in late March and April to the marshland. Spring ranges of collared individuals covered 16.8 km2, on average, significantly larger for males (25.6 km2) than for females (8.0 km2).

THE GENETIC CONSEQUENCES OF SOCIAL STRUCTURE: A SIMULATION APPROACH

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In nature most organisms occur in structured populations, which are rarely isolated but are part of a network of populations. Structure at the population level is taken into account in many population genetic studies and it is recognized as having consequences for genetic diversity, depending on processes such as the spatial distribution, habitat fragmentation, dispersal or migration patterns. However, in many species, populations are furthermore subdivided in social groups where organisms interact during most of their life time with related individuals. The types and complexities of social structures are enourmous and althought nature provides many examples of social organizations, this level of structure is usually ignored in population genetic studies. Instead, it is usually assumed that samples are obtained from populations at Hardy-Weinberg equilibrium. Empirical studies have found genetic substructure of populations due to the presence of social organization in different species. Specifically, these studies have found negative Fis values, a signal of deviations from random-mating, and have interpreted these results as an evidence of outbreeding. However, theoretical studies and simulation models have suggested that complex social structure combined with dispersal processes may generate similar patterns without the need to invoke inbreeding avoidance. Here we are interested in understanding i) what are the implications of departures from random mating conditions for population genetics studies; and ii) how the social structure influences the diversity and differentiation within and between groups. We have performed simulations under different types of social systems and different patterns of gene flow. We have quantified different genetic diversity statistics, namely, the Wright's F statistics and heterozygosities. Our results suggest that real data can be reproduced when social structures are taken into account in the modeling approach.

SPATIO-TEMPORAL CHANGES IN THE DISPERSAL PATTERNS OF TWO SYMPATRIC COLOBUS MONKEYS FROM CANTANHEZ NATIONAL PARK, GUINEA-BISSAU

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Western black-and-white colobus (Colobus polykomos) and Temminck's red colobus (Piliocolobus badius temminckii), often live in sympatry but exhibit contrasting social systems. C. polykomos live in small groups with one to three adult males, with mainly male-mediated dispersal but where females can also disperse. In contrast, P. b. temminckii live in large multi-male multi-female groups where dispersal is female-biased. To investigate whether genetic evidence supports the reported patterns of dispersal, we examined eight black-and-white and six P. b. temminckii social groups from Cantanhez National Park, Guinea-Bissau. Microsatellite markers revealed a lack of sex-biased dispersal for C. polykomos. Female-mediated gene flow better explained the genetic patterns found in P. b. temminckii, with evidence for less extensive male dispersal. In contrast, low mitochondrial diversity for the C. polykomos suggests that historical and/or long-range male-mediated gene flow might have been favored. In P. b. temminckii, the co-existence of three divergent mitochondrial lineages suggests that this population contains a secondary contact zone between divergent lineages. Female-biased dispersal may be a major factor contributing to the colonization by such differentiated mitochondrial lineages in the region. Overall, we found evidence for spatio-temporal change dispersal patterns of these colobus monkeys: mitochondrial DNA indicating historical and/ or long-range dispersal by mainly a single sex and microsatellite data suggesting that recently both sexes are dispersing within the population. This could indicate a local behavioral adaptation in response to changes in the environment that has becoming increasingly fragmented due to human-related activities. Ultimately, genetic data integrated in the study of dispersal patterns can provide great insights not only on the socio-genetic dynamics but also on the effect that anthropogenic disturbance may have on endangered populations.

FLUCTUATION OF SALINITY EFFECT ON THE COEXISTENCE OF TWO ROTIFER CRYPTIC SPECIES

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Salinity is an important factor affecting ecological and evolutionary processes in continental plankton. Brachionus plicatilis species complex is a group of planktonic rotifer cryptic species dwelling in salt and brackish waters. Two of the species in the complex (B. plicatilis and B. manjavacas) co-occur frequently in ponds across the Iberian Peninsula despite their great ecological similarity in terms of resource use and susceptibility to predation. These species widely overlap their salinity tolerance ranges, but have still different allocation patterns in diapause stages. These rotifers are cyclical parthenogens producing sexually diapause stages. As their populations are seasonally active, diapause is needed for their persistence. From observational data and theoretical work salinity fluctuation and diapause has been suggested to promote their coexistence. In order to test this hypothesis, B. plicatilis and B. manjavacas were grown together under several salinity fluctuation patterns in laboratory experiments simulating rows of growing seasons, so that the active populations in a growing season were founded from diapausing stages produced in the previous one. Our results reveal that high constant salinity brings to extinction to B. plicatilis, while low constant salinity has the opposite result. Under fluctuating salinity the species excluded was less consistent. The results can be used to design experiments in order to found the salinity fluctuation patterns required for coexistence.

A GENETIC APPROACH OF REEF COLONIZATION BY A SURGEON FISH (ACANTHURUS TRIOSTEGUS) IN LA RÉUNION ISLAND

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Resilience of coral reefs is highly dependent on the ability of reef organisms to colonize and survive, and thus to maintain these reefs connected. The aim of this study was to better understand, through population genetics, the fine-scale spatio-temporal colonization pattern of the surgeon fish A. triostequs. This species is well distributed all over the Indo-Pacific reefs and presents a good model to study the variability of reproductive success in marine organisms. The recruits, arriving in two locations 20 km apart, were collected from March 2011 to February 2013 using light traps. A total of 520 post-larvae were caught. Sampling was divided into several nested groups for each location: 4 reproduction seasons (mean duration: 3 months), 17 fishing campaign (6 days around the new moon), 64 days (whenever catches number was higher than 25 individuals). The genetic structure of these juveniles' populations was assessed using 14 microsatellites loci. For a given location, we found no significant genetic differentiation (1) between days among a fishing campaign, (2) between fishing campaign among a reproduction season, and (3) between reproduction season (maximum FST =0.0043, NS). On the 34 comparisons performed between locations over fishing campaign, only 4 were differentiated from the others (maximum FST = 0.012; P=0.042). Assignment tests did not allow identifying any genetic cluster over the overall sampling. These results showed the absence of temporal or spatial genetic heterogeneity between the post-larvae samples (no chaotic genetic patchiness): the pool of reproductive adults was stable through time. Moreover, the heterozygote deficiency (mean FIS = 0.153±0.006) can be linked to related fishes within samples. So, we performed sib-ship and half-ship reconstruction and found brothers and sisters among samples, showing that post-larvae migration is concerted. Key-words: colonization, A. triostegus, fish, population structure, microsatellites.

INFLUENCE OF STAGE SPECIFIC AVAILABILITY OF NUTRITIONAL RESOURCES ON THE DYNAMICS OF LABORATORY POPULATIONS OF DROSOPHILA MELANOGASTER

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The amount of nutritional resources available for individuals is known to play a major role in shaping the dynamics of a population. However, the effects of resource availability on the dynamics of stagestructured populations, remains relatively less explored. Here, we present our findings from a 49generation experiment on replicate populations of the common fruit fly, *Drosophila melanogaster*, subjected to four different combinations of larval and adult nutrition. We show that in general, increasing levels of larval nutrition stabilizes the dynamics, while increasing adult nutrition has the opposite effect. To study the dynamics in further detail, especially with the continuous change in the level of food available in the two life stages, we have constructed a stage-structured theoretical model for *Drosophila*. This model incorporates details of the various density-dependent feedback loops that determine the dynamics of *Drosophila* cultures. The model does a fair job in capturing both the qualitative and the quantitative nature of the dynamics of each of the four nutritional regimes studied empirically. Exploration of the model parameter space reveals that egg and adult dynamics behave very differently when subjected to different nutritional combinations. Insights obtained from this study have possible implications in managing biological populations with similar life-cycle.

GMETAPOP, A SOFTWARE FOR SIMULATING THE EVOLUTION OF SUBDIVIDED POPULATIONS IN SPECIES WITH COMPLEX DEMOGRAPHIC FEATURES

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Theoretical predictions of multilocus evolution in subdivided populations are usually very complex in realistic cases. Flexible simulation tools are thus needed to model the evolution of polygenic traits and their underlying genes in various selection or demographic scenarios. Several computer simulation tools have become available in recent years but none is user-friendly, as they need complex input files with large numbers of population genetics parameters. We propose here a simulation software that allows to model various key features («Life-history», «Demography», «Migration», «Selection», «Recombination») with a high level of complexity. We first combined two previous complementary versions of the program METAPOP: one with simple demographic features, but the possibility to model phenotypic values and selection, and one allowing more complex demography with overlapping generations. The new software (gMETAPOP) includes a Graphical User Interface (GUI) that greatly simplifies its use with no need to fill very large input parameter files so that users can focus on their biological model. Practically, the GUI is organised in different tabs (genome, phenotype-genotype modelling, demography, selection....), where the user simply chooses from different parameters' available options. Users are also guided by the GUI by being proposed default parameter values within possible ranges, thus avoiding trivial errors, and although a detailed helpfile is available, we have also summarized important parameter definitions uses with short statements (Tooltips). The simulation software is now further tested with a large range of simple modular tests, but also with tests based either on classical analytical predictions from population and quantitative evolutionary genetics or on published simulation results. These tests improve the efficiency for correcting bugs, and allow to assess the limits of the software. Various outputs are designed to be easily imported into R.

A COMPLETELY UNKNOWN LIFECYCLE IN MUSHROOMS: CYCLICAL INBREEDING AND HAPLO-DIPLOIDY

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Mycena galericulata (Basidiomycota, Agaricales) occurs in two forms, a clampless with two-spored basidia and a clamped with four-spored basidia. It is generally accepted that the two-spored form is haploid asexual (apomictic), and the four-spored form sexual (dikaryotic and heterothallic). In order to study the interrelationship between both forms, we performed mating tests and phylogenetic and genetic analyses of a sample of both forms. Surprisingly, our results are inconsistent with any currently known life-cycle. While the four-spored form is heterothallic indeed, we show that the two-spored form is diploid, and produces diploid spores via intra-tetrad selfing. However, the absence of genetic differentiation between both forms, and the high degree of heterozygosity in the two-spored form, indicate that the two-spored form frequently arises from the four-spored. We hypothesise that the two-spored form can again give rise to four-spored forms. Consistent with this, we discovered that a small percentage of fruiting bodies has both two-spored and four-spored basidia.

ECOLOGICAL DIVERGENCE BETWEEN LAKE-STREAM POPULATIONS OF AN EAST AFRICAN CICHLID FISH

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Divergent selection due to differences between environments often triggers ecological speciation. The cichlid species *Astatotilapia burtoni* occurs in the lacustrine environment as well as in the surrounding rivers of Lake Tanganyika. In this setting of replicated lake-stream populations, ecologically relevant traits such as gill rakers and the pharyngeal jaw apparatus (important for food uptake) are supposed to be under divergent selection as a consequence of different resource availabilities and utilization. Here, we study traits related to gill raker morphology (gill arch length, gill raker length and raker number), apply geometric morphometrics on pharyngeal jaws and analyse gut and stomach contents of four lake-stream systems in order to tests for shifts associated with differential foraging modes in *A. burtoni*.

MORPHOLOGICAL GENETIC DIVERSITY OF YAM (DIOSCOREA CAYENENSIS/D. ROTUNDATA COMPLEX) LOCAL VARIETIES IN BRAZIL

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Yams are one of the main roots and tuber crops in the world, especially within the species complex Dioscorea cavenensis Lam/D. rotundata Poir. The cultivation of yams is important both in traditional agriculture, and in family farming. Few studies have been conducted in Brazil with these species, including genetic diversity studies. As such, the objective of this study was to characterize the genetic diversity of yam (Dioscorea cayenensis / D. rotundata) local varieties from different regions in Brazil, using morphological characters. Thus, collections were made in several municipalities in the South, Southeast and Northeast regions. The tubers were planted in an experimental field, where they were evaluated for 18 morphological traits. These traits were transformed into binary data. The species identification of the accessions was carried out in accordance with the morphological evaluation performed. High genetic variability was observed and most of the variability was found between regions and between species, and this variability was spatially structured, with high and significant correlation between genetic and geographical distances. The cluster analysis and the principal coordinate analysis using the Jaccard index, showed a clear separation of accessions into two distinct groups: group I with accessions, identified as *D. rotundata*, originated from the Northeast and group II with accessions, mostly identified as D. cayenensis, originated in the Southeast region, while the accessions from the South were allocated either at an intermediate zone between the two groups, or included in either group. Given these data, one can infer that in Brazil there is a clear separation between D. cayenensis and D. rotundata species, and that *D. cayenensis* occurs mainly in the Southeast and South regions, and *D. rotundata* occurs predominantly in the Northeast region. Financial Support: FAPESP and CNPq.

DO NATIVE AND INVASIVE POPULATIONS OF OXALIS PES-CAPRAE DIFFER IN REPRODUCTIVE TRAITS?

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Biological invasions offer the opportunity to study evolutionary transitions in current timescales. Because reproductive strategies determine demographic and genetic characters of invasive populations, variations on reproductive characters have the potential to influence evolutionary processes during invasion, and the invasion process itself. Thus, comparative studies of reproductive systems between native and invaded ranges are crucial for understanding the mechanisms of plant invasion and for predicting microevolutionary changes in anthropogenic environments. *Oxalis pes-caprae* is a tristylous species native from South Africa and an invasive weed in Mediterranean climate regions worldwide. In its native habitat, the species presents three floral morphs, reproduces sexually and asexually, and has different cytotypes (2x, 4x, 5x). In most invaded areas, strong founder events lead to the introduction of the 5x short-styled morph only, leading to an exclusively asexual mode of reproduction. However, in the Mediterranean basin, new floral morphs and cytotypes with the ability to reproduce sexually were recently found. The aim of the present study was to assess evolutionary shifts in asexual and sexual reproduction traits between floral morphs and cytotypes, and between native and invasive populations of *O. pes-caprae*. For this, phenotypic characters directly linked with reproduction were assessed in plants from native and invasive populations growing in a common garden. The results of the sexual and asexual reproductive traits among floral morphs, cytotypes and invaded vs. native range are presented. The information gathered adds significant background information on the evolutionary biology and ecology of biological invasions. The knowledge of the probability and speed at which local adaptation evolves in invasive plants is particularly important for management practices, especially when evolutionary changes enhance ecological opportunities and invasive spread.

HABITAT CHOICE IN THE FLAT PERIWINKLE (*LITTORINA FABALIS*) IBERIAN ECOTYPES: AN EXPERIMENTAL APPROACH

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Although it is widely recognized that habitat choice can facilitate local adaptation, its contribution to speciation has been poorly studied. *Littorina* species (marine gastropods) offer unique models to study the mechanisms underlying ecotype differentiation. In the flat periwinkle *L. fabalis*, three ecotypes have been described in the Iberian Peninsula, each associated with different host algae/seagrass (*Fucus* sp., *Mastocarpus* sp., and *Zostera marina*). Whether this association results from consistent habitat preference/avoidance, represents the outcome of environmental selective pressures like predation and/or results simply from stochastic processes, has never been investigated. Here we conducted two series of laboratory experiments where we measured *L. fabalis* immediate habitat choice within a 30 minutes interval and long-term choice within a six days interval. Results were similar: while two of the ecotypes (living in *Fucus* sp. and *Mastocarpus* sp.) tend to choose their host algae more frequently than by chance, the opposite trend was observed for the third ecotype (living in *Zostera marina*). The

implications of these results will be discussed within the context of ecological speciation.

NOT EVERYBODY IS SOCIAL IN SOCIAL AMOEBAE: DICTYOSTELIUM DISCOIDEUM POPULATION PARTITIONING INTO AGGREGATING VS NON-AGGREGATING CELLS AND ITS EVOLUTIONARY SIGNIFICANCE IN FLUCTUATING ENVIRONMENTS

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The social amoeba *Dictyostelium discoideum* is widely known for its cooperative behavior. Upon starvation, millions of cells aggregate and form a fruiting body that allows better dispersion of dormant spores. But not all cells aggregate and take part in this social act. A part of the population ignores the aggregation signal and continues behaving as single cells, that can take advantage of incoming nutrients while the others are embedded in multicellular development. We have developed a new cell-tracking technique based on time-lapse fluorescence microscopy and image processing. This enabled us to quantify population partitioning into social/aggregating and unsocial/non-aggregating cells. We are currently investigating the effects of cell nutritional state, genetic background and environment structure on population partitioning. In addition, we propose a model to illustrate potential evolutionary benefits of population partitioning in fluctuating environments.

TEMPORAL GENETIC AND KIN STRUCTURE IN A BOREAL BIRD, THE SIBERIAN JAYS (PERISOREUS INFAUSTUS)

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Abundance of the Siberian Jay (*Perisoreus infaustus*) varies depending on weather condition during reproduction and predation. Siberian jays are territorial, long lived birds that form family groups with up to six individuals. The breeders are nepotistic towards their own kin while they display aggressive behavior towards non-kin. Relatively short dispersal distances and social behavior can lead to formation of kin clusters. We are combining observational data and microsatellite markers to examine temporal structure and genetic relatedness during a 15 year time period. Predation by the main predator, Goosehawks, lowered Siberian jay recruitment following high Willow grouse recruitment the year before. Furthermore, longevity of the Siberian jays dampens the effect of changes in temporal genetic structure during different population densities.

MALE-BIASED DISPERSAL PROMOTES LARGE SCALE GENE FLOW IN A SUBTERRANEAN ARMY ANT, DORYLUS (TYPHLOPONE) FULVUS

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Sex-biased dispersal is widespread in the animal kingdom and may strongly influence gene flow and population structure. Particularly army ants, important key-stone predators in tropical ecosystems, are prone to population fragmentation and isolation due to their extraordinary mating system: Queens are permanently wingless and propagate via colony fission and only the males disperse in mating flights. Here we report on sex-biased dispersal and the genetic population structure of an African subterranean army ant, *Dorylus (Typhlopone) fulvus.* Using maternally inherited mtDNA and bi-parentally inherited microsatellites we found strong geographical structuring of mtDNA haplotypes, whereas the microsatellite genetic population structure was less pronounced. Strong mtDNA, but significantly lower microsatellite genetic differentiation translated to a more than an order of magnitude larger male migration rate compared to that of queens. This reflects the low motility of queens and strong, promiscuous dispersal by males. Thus, the well flying *D. fulvus* males appear to be the sex to promote large scale gene flow. With this study we aim to achieve a better understanding of how the evolution of sex specific dispersal patterns and mating systems affect the population structure and phylogeography of species.

EFFECT OF CROWDING ON HATCHING STRATEGIES OF TWO CRYPTIC SPECIES OF CYCLICAL PARTHENOGENS

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Monogonont rotifers are cyclical parthenogens combining asexual reproduction with occasional bouts of sexual reproduction. This life-cycle is considered an adaption to time-varying habitats as the asexual phase allows clonal propagation in the water column before harsh conditions begin, while sexual reproduction makes re-colonization possible when good conditions are restored. The sexually-produced diapause egg can resist periods of habitat unsuitability (desiccation, intense competition, presence of predators) and is induced as a response to any environmental cue (abiotic or biotic) that anticipates the advent of unfavorable conditions. Similarly, it is crucial that dormant eggs should be able to detect such conditions before hatching. The beginning and end of diapause are thus two important transition points in their life cycle, whose success is dependent on their timing. In the extensively studied genus Brachionus, sexual reproduction is density-dependent, and induced by a chemical signal (mixis induction protein) produced and released into the environment by the rotifers. We believe that the same protein can influence hatching because as population density increases, the probability of success of one hatchling against a high density of clonally multiplied competitors decreases. Aiming at testing this hypothesis, we investigate the influence of population density on diapausing egg hatching of two rotifer cryptic species. Brachionus plicatilis and B. manjavacas belong to a species complex of planktonic rotifers dwelling in temporary waters which frequently co-occur in ponds across the Iberian Peninsula. We test whether rotifer diapausing eggs can detect infochemicals during dormancy and assess the presence of conspecific and intraspecific high population densities and if such an assessment results in changes in hatching strategies.

FREQUENT RECOMBINATION SHAPES THE EPIDEMIC POPULATION STRUCTURE OF A FRESHWATER CYANOBACTERIUM

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Based on increasing knowledge, cyanobacteria are prone to gene-flow, as are other prokaryots. The planktonic genus *Planktothrix* showed remarkable signals of both homologous and non-homologous recombination. However, it is not yet clear how frequent recombination is and whether this phenomenon impacts population structuring. In the frame of a population genetics study, we characterized 290 Planktothrix strains isolated from seven neighbouring lakes in the subalpine Italian region with multi locus sequence typing. Four of six house-keeping loci analysed were polymorphic, resulting in 20 distinct multi-locus genotypes. Association indices among alleles at different loci were suggestive of a so called 'epidemic population structure', thus intermediate between clonal and panmictic structures. Further analyses carried out by using the software ClonalFrame, which is able to infer genealogies from multy-locus datasets impacted by recombination, supported this view by detecting: i) three major clades affected by three distinct recombination events, ii) a recombination rate about equal to the mutation rate and iii) the fact that recombination had an impact in introducing molecular diversity more than double the mutation rate. Furthermore, among-lake diversity, determined by the analysis of molecular variance and verified over an annual cycle in three of seven lakes, appeared as influenced by both recombination processes and local clonal expansions. Our observations suggest that microevolution of the cyanobacterium *Planktothrix* is frequently impacted by recombination, thus determining a reliable epidemic population structure.

GOOD TIMES BAD TIMES: SHOULD I STAY OR SHOULD I GO?

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The classical theory of dispersal predicts that heterogeneity in local density may select for the evolution of density-dependent dispersal strategies. In this case, individuals should disperse away from highdensity habitats, i.e. positive density-dependent dispersal, thus increasing their chances of breeding in favourable low-density habitats. Various empirical studies support this view, while others suggest the opposite, i.e. negative density-dependent dispersal. Using a theoretical model, we show that temporal variation in habitat density may be a key factor in explaining these contrasting observations. Stable environments tend to promote the evolution of negative density-dependent dispersal rates, whereas unstable environments tend to promote the evolution of positive density-dependent dispersal rates. We also show that temporal variation in habitat density greatly affects the evolution of unconditional dispersal strategies, with unstable environments typically favouring the evolution of higher unconditional dispersal rates.

THE INTERACTION OF SACCHAROMYCES PARADOXUS WITH ITS NATURAL COMPETITORS ON OAK BARK

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Saccharomyces cerevisiae is a well-studied laboratory model organism but its natural history is poorly understood, and confounded by domestication. Its close relative, *S. paradoxus*, is often found on oak trees. *Saccharomyces* species seem to be adapted for rapid growth in sugar rich environments (like the grape juice that is fermented for wine production wine), so we wanted to study how they grow in their natural, but very different, environment of oak bark. Oak tree *S. paradoxus* grows well on oak bark medium that is sterilised by heat, filtration, or chemically. But its growth is almost completely suppressed when the microbes that normally inhabit the oak bark are allow to compete. Using a combination of culturing, oak bark competition experiments, and metagenomic sequencing we are investigating what the most important natural competitors of yeast are in its natural habitat.

TEMPORAL POPULATION GENETIC PATTERNS IN A POLYCHAETE WITH MULTIPLE LARVAL DEVELOPMENTAL MODES

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Many marine invertebrates have complex life cycles in which in one or more larval stages precede the adult stage. The larval developmental mode of a species can affect population dynamics through differences in fecundity, larval mortality and dispersal potential. Developmental mode may also affect temporal stability of populations. Due to multiple risks that planktonic larvae face during the pelagic stage, benthic species with planktonic larvae may be more vulnerable to temporal variation in population genetic structure, whereas species with benthic or non-dispersive larvae are expected to be temporally more stable. We examined temporal genetic patterns in populations of a poecilogonous polychaete with within-species variation in developmental mode. *Pygospio elegans* can produce small free swimming larvae feeding in the plankton, or benthic larvae feeding on nurse eggs while brooded in capsules in the maternal tube. With our microsatellite analyses, we found low but significant temporal genetic differentiation in the populations with high frequency of benthic larvae. This pattern is likely to have been caused by genetic drift, since estimations of population density, Ne and migration rates among these populations were low. Stable temporal genetic structure and higher Ne was observed in the strictly planktonic population. In one population (Dutch mudflat with both larval modes), a distinct genetic turnover was detected among the temporal samples, which could have been caused by habitat change (due to harsh winter) and (re)colonization from neighboring planktonic populations. Over a long time scale, metapopulation dynamic patterns may be likely in *P. elegans*, especially in planktonic populations in high disturbance intertidal habitats. Because of the observed polymorphism in developmental mode seen in *P. elegans*, it is a good model species for research on the consequences of life history variation on population genetic patterns.

TEMPORAL AND SPATIAL VARIATION IN PREVALENCE OF THE PARASITE SYNGAMUS TRACHEA IN A METAPOPULATION OF HOUSE SPARROWS (PASSER DOMESTICUS)

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When investigating parasite-host dynamics in wild populations, a fundamental parameter to investigate is prevalence. This quantifies the percentage of individuals infected in the population. Investigating how prevalence changes over time and space can reveal interesting aspects in the parasite-host relationship in natural populations. We investigated the dynamic between a common avian parasite (Syngamus trachea) in a host metapopulation of House Sparrows (Passer domesticus) on the coast of Helgeland in northern Norway. We found that parasite prevalence varied in both time and space. In addition, the parasite prevalence was found to be different between demographic groups in the local populations. Our results reveal just how complex the dynamic between a host and its parasite may become in a fragmented landscape. Although temperature may be an important factor, the specific mechanisms causing this complexity are not fully understood, but needs to be further examined to understand how parasite-host interactions may affect the ecological and evolutionary dynamics and viability of host populations.

SEX-SPECIFIC FITNESS CONSEQUENCES OF TRANSLOCATION IN HOUSE SPARROWS

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Translocations are increasingly used in conservation programmes to reinforce small populations. Success of translocations in relation to characteristics of the species, the location or the translocation event, such as propagule size, is well studied. However, much less is known about individual variation in genetic contribution to the release population. Here, we compare sex-specific fitness of resident and introduced individuals based on genetic parentage, following a translocation experiment in house sparrows. The genetic mating pattern revealed assortative mating based on origin, where introduced males were highly unlikely to mate with resident females. Introduced males had fewer mates and fledglings than resident males. Origin had no effect on number of fledglings in females. The effect of origin on number of fledglings was sex-specific as indicated by the significant interaction between sex and origin. The number of recruits and total annual fitness contribution did not differ between resident and introduced individuals. In the house sparrow, male badge size has a function in intra- and intersexual selection. It is likely that the influence of badge size is of greater importance for introduced males, which have to compete with resident males over nest sites and mates. However, the relationship between number of mates and badge size did not differ between resident and introduced males. Still, the effect of badge size on number of fledglings was stronger in introduced males. Our results provide insights about sex-specific individual heterogeneity in reproductive success of introduced individuals, and the potential role of sexually selected traits for the genetic consequences of a translocation event.

FINE-SCALE POPULATION STRUCTURE OF HUMPBACK WHALES IN SOUTH AFRICA COAST

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Humpback whales (Megaptera novaeangliae) undertake extensive seasonal migrations from polar feeding grounds in the summer to tropical breeding grounds in winter, displaying high fidelity to the same breeding and foraging areas. En route between these areas, some humpback whales utilize the coastal waters of southern hemisphere continents as migratory corridors. On South African coast two distinct migratory corridors of humpback whales are present. One takes humpback whales past the west coast of South Africa in autumn, en route to breeding grounds off Gabon, Congo, Angola and coastal and offshore areas in Gulf of Guinea. And the other takes them past the east coast of South Africa, en route to coastal waters of Mozambique and Madagascar. The aim of the present work was to assess a fine-scale population structure of humpback whales on the two migratory paths in the South African coast, combining mitochondrial DNA and microsatellites markers in order to characterize individual and population levels of genetic structure variation in this region. It was amplified, sexed, genotyped and sequenced a total of 484 samples from west and east coast. The results revealed population differentiation and restricted connectivity of humpback whales from west and east coast of South Africa. Differences on fidelity and seasonality to each region were also found, with humpback whales from west coast showing high levels of long term fidelity to the area, with identified animals returning to the same area in multiple years and between long periods of time and broader seasonality with

several animals been sighted and resighted in summer months suggesting that this region besides functioning as a migratory corridor serve as an important feeding ground to some animals of the population. A better understanding of the scale of this behaviour off the west coast of South Africa and the importance of this area for the breeding ground of Gulf of Guinea should be achieved in the future.

Symposium

34. General Symposium

24 August



Program

Saturday 24 August

Session(s): 14

D24SY34RT10:30R9

ECO-EVOLUTIONARY DYNAMICS AND THE EVOLUTION OF SEX

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The evolution of sexual reproduction is one of the most important and controversial problems in evolutionary biology because sexual reproduction is almost universal while its inherent costs have made its maintenance difficult to explain. Major hypotheses on the evolution of sex predict sex to be advantageous when the environment changes frequently over time. Recurrent change can occur as a result of eco-evolutionary feedback dynamics but has not been considered in the context of the evolution of sex. An eco-evolutionary feedback loop occurs when environmental change causes natural selection in a population within a few generations, and the resulting trait evolution then modifies the environment, causing further selection and evolution, and so on. An example for this eco-evolutionary feedback comes from a predator-prey system where the algal prey evolves a defence against predation when grazing by rotifer is intense, and loses the defence, but gains competitive ability when the predators (rotifers) are scarce and prey are abundant. I present experimental results showing that the rate of sex evolved to higher rates in the rotifer when eco-evolutionary feedback dynamics occur. In contrast, the rate of sex evolved to lower rates in control populations where the trait evolution in the algae and thus the eco-evolutionary feedback was prohibited. Thus the changes driven by the interplay of ecological and evolutionary change on one time scale can provide conditions allowing for a more globally relevant explanation for the evolution of sex.

D24SY34RT10:54R9

BACTERIAL SEX CAN ENHANCE COOPERATION

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Mobile genetic elements, such as plasmids, are the most prominent mechanism of bacterial sex and play a large role in bacterial adaptation. Plasmids move between bacteria by conjugation, an active, unidirectional horizontal transfer of genetic material from donor to recipient cells. Since conjugation is costly for donors and is primarily controlled by the plasmid itself, horizontal transfer is usually seen as parasitism of the bacterial hosts. However, chromosomal genes can also influence horizontal transfer. Transferring plasmids could benefit the donor host by generating a specific behavior in recipients. Using both modeling and experimental work we show that if cooperation genes are located on the plasmid, conjugation can benefit the host bacteria by facilitating the maintenance of cooperation. We modeled populations of donor and recipients and showed that horizontal transfer increases the selection for cooperation by increasing relatedness and the overall amount of public good secreted in the population. This effect in turn allows for the selection of transfer: when cooperation is needed, genes increasing donor and recipient abilities can be selected both on the plasmid and on the chromosome. Our results could explain the high frequency of genes related to cooperation that are located on plasmids. In addition to the models, we used a synthetic system with independent control of cooperation and conjugation to experimentally test their relationship in Escherichia coli. We introduced multiple fluorescent proteins into bacterial strains and were able to measure the frequency of different types of bacteria, plasmids, as well as transfer events, by flow cytometry. We show that plasmid transfer indeed increases the selection for cooperation. Based on our models and experiments, we can conclude that the role that bacterial sex plays in evolution is strongly linked to the presence of and interaction with cooperation via public good secretion.

D24SY34RT11:18R9

IS SEXUAL SELECTION INVOLVED IN THE MAINTENANCE OF THE COOPERATIVE MOUND-BUILDING IN *MUS SPICILEGUS*?

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The mound-building mouse (*Mus spicilegus*) possesses a unique cooperative behaviour amongst mice. In the beginning of autumn, groups of several individuals construct large earthen mounds in which they overwinter. This building is partly explained by kin selection. However, the presence of some distant or unrelated individuals suggests that kin selection may not suffice to explain this cooperative behaviour. Some observations in animal species suggest that cooperative behaviour increases access to mates. Investigations into whether sexual selection is involved could therefore provide significant insight. Here, spontaneous cooperativeness, exhibited during mound-building in captivity, was first assessed in a sample of 30 male mice captured in the field in Bulgaria. Then, we tested whether male cooperativeness influences mate preference of 20 females captured in the same area. Females significantly preferred cooperative males over non-cooperative ones. This is the first empirical evidence for the influence of sexual selection on cooperative behaviour in another species than humans.

D24SY34RT11:42R9

CONDITION AND MOLECULAR MATING STRATEGIES IN DROSOPHILA MELANOGASTER: LARGE FEMALES GET MORE, SMALL MALES TRY HARDER

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Theory predicts that males in good condition should produce larger sexually selected traits, such as ejaculates, but the allocation of ejaculates should also vary with the number of mating opportunities available to males. Fruit flies, *Drosophila melanogaster*, who develop under crowded larval conditions have reduced body size, and these small males posses fewer ejaculate resources. We show, however, that these small males transfer relatively higher proportion of their reserves of a key seminal protein, sex peptide, at mating, and manage to match large males in the quantity they transfer to females. Small males could potentially benefit form this strategy because they have fewer mating opportunities: thus they are predicted to invest relatively more in each mating opportunity they get. Large females receive larger quantities of the receptivity-inhibiting sex peptide from males of all sizes, but unexpectedly these large females remate sooner than small females. Thus, female mating frequency is strongly dependent on the female developmental environment. Future work should aim to uncover to what extent female condition and male influences interact to regulate female remating rate.

POSTERS

D20SY34PS0520

FEWER INVITED TALKS BY WOMEN IN EVOLUTIONARY BIOLOGY: MEN ACCEPT INVITATIONS TO SPEAK MORE OFTEN THAN WOMEN

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Lower 'visibility' of female scientists, compared to male scientists, is a potential reason for the under-

representation of women among senior academic ranks. Visibility in the scientific community stems partly from presenting research as an invited speaker at organised meetings. We analysed the sex ratio of presenters at the European Society for Evolutionary Biology Congress 2011, where all abstract submissions were accepted for presentation. Women were under-represented among invited speakers at symposia (15% women) compared to all presenters (46%), regular oral presenters (41%) and plenary speakers (25%). At the ESEB congresses in 2001–2011, 8–23% of invited speakers were women. This under-representation of women is partly attributable to a larger proportion of women, than men, declining invitations: in 2011, 50% of women declined an invitation to speak compared to 26% of men. We expect invited speakers to be senior scientists or authors of recent papers in high-impact journals. Considering all invited speakers (including declined invitations), 23% were women. This was lower than the baseline sex ratios of early–mid career stage scientists, but was similar to senior scientists and authors published in high-impact journals. High-quality science by women therefore has low exposure at international meetings, which will constrain Evolutionary Biology from reaching its full potential. We wish to highlight the wider implications of turning down invitations to speak. In particular, underrepresentation of women among invited speakers reduces the number of female role models for evolutionary biology students and contributes to the leaky pipeline. We encourage conference organisers to implement steps to increase acceptance rates of invited talks.

VARIATION OF ANAL FIN EGG-SPOTS ALONG AN ENVIRONMENTAL GRADIENT IN A HAPLOCHROMINE CICHLID FISH

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Color and pigmentation patterns seem to play a central role in the explosive radiations of cichlid fish species in the East African Great Lakes. The majority of African cichlid species are members of a single tribe, the Haplochromini. A characteristic feature and possibly a key-innovation of haplochromine cichlids are the so-called egg-spots in form of ovoid markings on the anal fins, made up of carotenoid based pigment cells. Egg-spots show a tremendous variability among different haplochromine species with regard to number, size, intensity, coloration and position on the fin. In this study we investigate the differentiation of egg-spots within a single species, *Astatotilapia burtoni*, which inhabits lacustrine and riverine environments of the Lake Tanganyika drainage. This phylogeographic setting enables us to study egg-spots in different habitats under divergent selection regimes. Four lake-stream population groups were analyzed and differences between stream and lake populations as well as between sexes were detected, indicating that natural and sexual selection influence the egg-spot phenotype in this species.

QUANTIFYING SEX ROLES DURING INCUBATION IN SONGBIRDS: A COMPARATIVE ANALYSIS

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Parental care, if present, is provided either by one sex or by both sexes. In birds, an important component of parental care is incubation of eggs. In uniparental incubators (only one sex is responsible for incubation), females usually incubate. As incubation behavior is time-consuming and energetically demanding, there might be an upper limit on incubation effort in these uniparental incubators. To overcome this limit and increase overall incubation intensity, males might help either directly by incubating the eggs (complementary parental care) or indirectly by feeding the incubating female on the nest (task specialization). To reveal whether there is an upper limit on incubation intensity in female-only incubators, we compared average nest attentiveness (% of daytime spent on eggs) in female-only incubators with species where only females incubate but males feed them, and with species where both sexes incubate. Further, we studied how total nest attentiveness changed with the degree of male help. Our phylogeny-based comparative analyses were based on a sample of 320 species of songbirds and data extracted from primary literature. We confirmed that i) total nest attentiveness increased from female-only incubators through species where incubating females are fed by males to biparental incubators. Further, we showed that ii) in species where females incubate and males feed them, nest attentiveness increased with the intensity of male incubation feeding, and that iii) in biparentally incubating species, male nest attentiveness correlated positively with total nest attentiveness, whereas female attentiveness did not. These results demonstrate that male help is indispensable for increasing total incubation effort in songbirds, which might be in turn dictated by environmental selective factors (e.g. temperature, nest mortality).

NUCLEOSOME POSITIONS IN EUKARYOTES AND ARCHAEA PRIMARILY REFLECT RATHER THAN AFFECT SEQUENCE EVOLUTION

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Histone proteins can affect the evolution of the sequences they bind. Their presence can promote or impede the formation of DNA lesions or interfere with efficient DNA repair, so that some mutations may occur more, others less frequently in a nucleosomal context. In addition, selection can eliminate mutations that disrupt beneficial nucleosome architecture, for example around promoters. Conversely, evolution at the sequence level can affect chromatin organization. Notably, as nucleosomes form preferentially on more bendable DNA, mutations that render the DNA template less bendable can prompt the nucleosome to shift to a position that is more conducive to nucleosome formation. In short, nucleosomes (and other DNA-binding proteins) can both affect and reflect sequence evolution. Several studies, screening a variety of eukaryotic genomes, have observed non-random associations between patterns of sequence evolution and nucleosome footprints. But do these associations reflect mutational biases or selection or are they evidence for sequence-directed nucleosome repositioning through evolution? And can we use comparative genomic data to discriminate between these competing hypotheses? Combining large-scale phylogenetic reconstruction of substitution histories with highresolution nucleosome maps from eukaryotes and archaea, we show that - at the level of betweenspecies divergence - substitution dynamics around nucleosome dyads are not consistent with mutational biases or purifying selection. Instead, they are consistent with frequent local nucleosome repositioning through evolution. Our results highlight the importance of considering the direction of causality between genetic and epigenetic change when interpreting variation at the genetic or epigenetic level and especially when trying to infer selection from patterns of genetic conservation.

34. General Symposium

D23SY34PS0147

THE EVOLUTION OF PUNISHMENT IN PUBLIC GOODS GAMES

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The evolution of costly cooperation, i.e. helping others at a cost and with no guarantee of future returns, is a challenge. Mathematical and computational models as well as experiments show that one way to stabilize cooperation between selfish actors is to allow them to punish each other, so called peer punishment. While such punishment seems to be relatively stable, it is not trivial to explain its evolution. One solution for the evolution of peer punishment, voluntary participation, works in a similar way for pool punishment, where the decision to punish is made before the public goods game. If participation is not mandatory, antisocial punishment should be rare. Both mathematical models and behavioural experiments are discussed.

SPECIES RECOGNITION IN TWO SYMPATRIC EUROPEAN CICADAS OF GENUS TETTIGETTALNA (HEMIPTERA, CICADIDAE): IS ACOUSTIC BEHAVIOUR ENOUGH?

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The genus *Tettiqettalna* is composed of nine morphologically similar cicada species, being seven of these endemic to the southern Iberian Peninsula. Males produce calling songs to attract females for pair formation and reproduction. These acoustic signals are thought to be species-specific mate recognition systems with an important role in the maintenance of reproductive isolation among cicadas. Because some Tettigettalna species are often found in sympatry, we investigated the patterns of intra- and interspecific variation in male calling songs of both allopatric and sympatric populations of two sibling species, T. argentata and T. mariae. Morphometric and genetic analyses were also performed. The results confirmed the distinction of species-specific calling songs based on time variables, but the calling songs of both species recorded in sympatry were downshifted in sound frequency. Interspecific variation in morphology is low but both species revealed a significant increase in general body size in sympatric populations. Sequence analyses with one mitochondrial gene (cytochrome oxidase I) and two nuclear genes (calmodulin and elongation factor) revealed that T. argentata is geographically structured, forming two distinct clades (North and South) and that specimens of T. mariae cannot be entirely distinguished from those of *T. argentata* (clade South). The genetic diversity and the distribution range are much smaller for T. mariae than for T. argentata and most mitochondrial haplotypes detected in *T. mariae* are shared with *T. argentata* (clade South), which is usually found in sympatry and parapatry with *T. mariae*. The genetic overlap between this pair of sibling species might have been caused by incomplete lineage sorting or recent introgression. It remains unclear whether the differences in species-specific male songs are enough to fully prevent hybridization when these species are in sympatry.

POLYMORPHISM AT FROST GENE EXPLAINS DEVELOPMENTAL TIME VARIABILITY IN DROSOPHILA AMERICANA

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Developmental time (DT) is a variable and complex trait that is of great relevance to all organisms. Nevertheless, in Drosophila, the specific genes contributing to DT variation in natural populations remain largely unknown. Here, we make the first attempt to characterize the molecular basis of DT variation in Drosophila americana, a species of the virilis group of Drosophila that is distantly related to D. melanogaster. The comparison of divergent species can shed light on the generality of the molecular basis of DT differences. For this purpose, we developed a set of 43 indel markers equally spaced along the D. americana genome and conducted two F2 association experiments. Associations are found between markers in all chromosomes and DT but two regions stand out, namely, a region in the middle of Muller's element C, and the Frost (Fst) gene region on Muller's element E. Variation at the Frost gene explains 25.3 % of the variation in DT in a sample of unrelated individuals from the same population. Fst is known to be involved in the Drosophila cold response, but there is a growing body of evidence suggesting that it is also a developmental gene.

THE SCENT OF INBREEDING: A MALE SEX PHEROMONE BETRAYS INBRED MALES

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Inbreeding depression results from mating among genetically related individuals and impairs reproductive success. The decrease in male mating success is usually attributed to an impact on multiple fitness-related traits that reduce the general condition of inbred males. However, the reduced mating success of inbred males could also be a consequence of strong selection on females to avoid mating with an inbred male. This is especially relevant if females gain direct benefits through appropriate mate choice (parental care, territory defences, nuptial gifts) or if they suffer direct costs should they mate with an inbred male (decreased offspring viability and fertility). Here we find that the production of the male sex pheromone is reduced significantly by inbreeding in the butterfly *Bicyclus anynana*. Other traits indicative of the general condition, including flight performance, are also negatively affected in male butterflies by inbreeding. Yet we unambiguously show that only the production of male pheromones affects mating success. Thus, this pheromone signal informs females about the inbreeding status of their mating partners. We also identify the specific chemical component, hexadecanal, likely responsible for the decrease in male mating success. Our results advocate giving increased attention to olfactory communication as a major causal factor of mate-choice decisions and sexual selection.

REPRODUCTIVE BIOLOGY OF THE 'ZYGOPETALUM MACULATUM' ORCHID COMPLEX AND ITS CONSEQUENCES FOR SPECIES DIVERSIFICATION

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Reproductive and pollination biology studies are crucial to understand lineage diversification and thus species delimitation in plants. The 'Zygopetalum maculatum' complex comprise a taxonomically challenging species complex restricted to the 'campos de altitude' (highland fields) in the south and southeastern region of Brazil. Although reproductive isolation may be one of the most important proprieties of species divergence, there are no studies available for this group. We studied phenology, floral biology, pollination and breeding systems of three species of 'Z. maculatum' complex, Z. brachypetalum, Z. intermedium and Z. triste. We observed a main flowering peak in the dry and cold season, when few rewarding plants are in bloom. Pollination is deceptive, rare and achieved by Bombus atratus (Apidae) in the beginning of the flowering season. Of the 68% individuals that produced inflorescences only 0.8% developed fruits to completion. We found no difference in the number of fruits formed from auto-pollination and cross-pollination (within/between individuals and between species) and the number of viable seeds. About 30% of seeds are polyembrionic, which suggests the occurrence of sporophytic apomixis. As for other deceptive pollinated orchids, reproductive success is pollinator-limited. Deposition of pollinia in the stigmatic cavity is necessary for fruit formation and spontaneous self-pollination is extremely rare. This scenario rises the hypothesis that allopolyploidy apomixis evolved as a response to increase colonization ability in the harsh environment of 'campos de altitude' and not as an alternative strategy to avoid inbreeding caused by self-compatibility. Phylogeographical and population genetic studies are in progress to understand whether the low reproductive success determined by deceptive pollination is enough to maintain heterozygosity in populations and the role of apomixis in the diversification of this group.

MAINTENANCE OF CO-OPERATION BY POSITIVE FEEDBACK IN QUORUM SENSING NETWORKS

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Signal blind mutants are naturally occurring bacterial strains that are defective for cell to cell signalling. These mutants can produce signal but do not respond to it, so they have reduced expression of quorum sensing regulated genes. Public goods are often regulated by quorum sensing and so signal blind mutants act as defectors, exploiting signal competent co-operators. Critically, signal blind strains are unable to increase their signal output in line with co-operators, because signal production is increased in response to signal, via a positive feedback loop. Therefore signal concentration in the environment is expected to be reduced when signal blind defectors are common. When public goods are regulated by quorum sensing, lower signal concentrations may reduce the production of public goods, minimising exploitation. Using knock-out strains of *Pseudomonas aeruginosa* we demonstrate that signal in the environment decreases as signal competent co-operators become rare. We show that this translates to a reduced co-operator output of secreted protease. Using competition experiments we then demonstrate that this can lead to negative frequency dependence of co-operator fitness, because co-operators behave as phenotypic defectors when rare. Frequency dependence has important implications as socio-microbiology is applied to new fields, notably the emergence of drug resistance.

ENGINEERING MICROSATELLITE MARKERS TO STUDY AND COMPARE A WIDE RANGE OF SPECIES

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We have developed a set of conserved avian markers with high cross-species utility in order to save resources and enable new comparisons between species. These markers will not only reduce the necessity and expense of microsatellite isolation for a wide range of genetic studies, including avian parentage and population analyses, but will also now enable comparisons of genetic diversity among different species (and populations) at the same set of loci, with no or reduced bias. No other marker type enables such comparisons. Additionally, these markers (i.e. microsatellites), in contrast to single nucleotide polymorphism (SNP) and genotyping-by-sequencing methods, are readily typed in samples of low DNA quality or concentration (e.g. non-invasive samples or museum specimens), and enable the quick cheap identification of species, hybrids, clones and ploidy. We selected zebra finch (Taeniopygia guttata) sequences that possessed a repeat region and that displayed high sequence similarity to chicken (Gallus gallus). Each primer sequence was a complete match to zebra finch and, after accounting for degenerate bases, at least 86% similar to chicken. These markers are of especially high utility in passerines, but also show utility in non-passerine species. The approach used here can be applied to other taxa in which appropriate genome sequences are available.

POLLINATOR SELECTION ON FLORAL TRAITS IN A NATURAL HYBRID ZONE OF TWO GENERALIST PLANT SPECIES

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Hybridization can create natural hybrid zones where two related species mate and produce (some) viable hybrids. These areas are natural laboratories to investigate evolutionary processes and mechanisms operating in the organisms integrating them, especially gene flow and selective pressures on phenotypic traits. For example, hybrid zones often present a striking profusion of flower morphologies and floral traits may profoundly influence both formation and fitness of hybrids primarily through the effect on pollinator behaviour. Within the Circum-Mediterranean genus Anacyclus, the rayed-head species A. clavatus co-exists with the rayless-head A. valentinus along the Western Mediterranean coast. In these mixed populations, a high phenotypic diversity in the number and size of ray florets has been observed revealing the existence of a dynamic hybrid zone. The presence of rays has been shown to have significant consequences on pollination, primarily enhancing the attractiveness of heads and consequently influencing the levels of outcrossing. These species show a generalist pollination syndrome since they are commonly visited by more than 80 species of insects. However, the role of pollinator preferences in hybrid zones between highly generalist species remains unknown and its study might help understanding how generalist interactions exert selection of floral phenotypes. Hence, this hybrid zone provides an exceptional microevolutive framework to explore how generalist pollinators are driving the evolution of floral phenotypes. To explore the role of pollinator preferences on floral traits we investigated floral trait variation on a natural hybrid zone of A. clavatus and *A. valentinus* and assessed the number and type of pollinator visits on each phenotype. Furthermore, we performed phenotypic manipulations of floral morphologies for a better understanding of pollinator preferences

HSP70 GENE FAMILY EVOLUTION AND DIFFERENTIAL EXPRESSION IN MICROBIAL EUKARYOTES

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In eukaryotes, members of the 70 kDa heat-shock protein family (Hsp70) are separated into four subfamilies according to their sub-cellular localisation and function. While in metazoans multiple cytosolic Hsp70s can be found, which are either constitutively expressed and/or stress-inducible, information about the copy number and diversity as well as differential expression of *hsp70* genes in microbial eukaryotes is scarce. In this study, we have characterised the *hsp70* gene family of the ciliate *Paramecium caudatum* to gain insight into the evolution and differential response to temperature stress of the distinct Hsp70 family members in protists. We further focused on the evolution of heatinducibility in cytosolic *hsp70s* and investigated intraspecific differences in *hsp70* gene expression to evaluate its potential use as biomarkers for temperature adaptation studies in Paramecium. Phylogenetic analyses disclosed five homologous groups, each with a closer relationship to orthologous hsp70s of Paramecium tetraurelia than to another P. caudatum Hsp70-group, indicating duplication events preceding Paramecium speciation. Furthermore, heat-shock expression studies and comparative EST analyses revealed one cytosolic group as the major heat inducible *hsp70s* in both *P*. caudatum and P. tetraurelia, suggesting a functionally conserved evolution of this gene family in *Paramecium*. In addition, the herein developed RT-qPCR assay unveiled different expression patterns between diverse thermal tolerant P. caudatum clones, demonstrating the potential use of Hsp70 as a biomarker for environmental stress studies in *Paramecium*. Interestingly, our analyses also suggest that heat-inducibility of cytosolic *hsp70s* evolved several times independently during the course of eukaryotic evolution, thereby indicating convergent evolution during Hsp70 subfunctionalization.

SEX AND COOPERATION IN DIGITAL COMMUNITIES

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The evolution of cooperation and the evolution of sex remain two of the most widely debated evolutionary topics, affecting communities of species ranging from microorganisms to humans. Both present a similar dilemma of evolving and maintaining a trait with long-term benefit and short-term cost, and there is no unifying theory for either. It has been suggested that plasmid conjugation, a form of bacterial sex, promotes cooperation based on secretion of public good molecules. Here we propose that the relationship between conjugation and secretion goes in both directions: plasmid transfer may promote cooperation by spreading the cooperating plasmid, but also, if cooperation is selected for, it may in turn promote sex, as an effective mechanism for its maintenance. In short, sex affects cooperation, but cooperation may also affect sex. To test these theories, we used an in silico system, Aevol, in which large populations of digital organisms compete, mutate and evolve over tens of thousands of generations. Digital individuals may evolve their plasmid donor and recipient abilities as well as the level of secretion of the costly, diffusible public good molecule. Results of our experiments show that transfer was more likely to evolve when secretion was present than when individuals could not secrete. It was primarily the donor ability that increased, while only in a handful of cases the recipient ability decreased, making individuals more immune to plasmid infection. However, the secretion genes were not always located on the plasmid, as we expected. Instead, especially when the transfer rate evolved to high values, it was the metabolic genes that were primarily encoded on the plasmid, turning it into a kind of a selfish genetic element. In conclusion, we demonstrate that in the diverse communities of digital organisms, individuals engage in complex relationships relying on costs and benefits of both cooperation and sex that in turn shape their evolutionary fate.

TESTING THE ROLE OF REPRODUCTIVE PROTEINS IN THE ECOLOGICAL SPECIATION OF *LITTORINA* SPECIES

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The genus *Littorina* is a well known model for studying ecological speciation as non-allopatric divergence across the intertidal zone has been observed in several species from this genus. Although reproductive proteins (RPs), specially those involved in mate recognition, have been shown to evolve rapidly under positive selection, in both internal and external fertilizers, and to cause assortative mating within populations, the contribution of RPs to the speciation processes observed in this genus is unknown. With the present project we aim to study how sexual selection at RPs contributes to ecological speciation observed in *Littorina*. To identify RPs expressed in this genus we sequenced the transcriptome of non-reproductive tissues (foot and head), as well as male and female reproductive tissues (prostate, seminal vesicle, testis, ovary and bursa) of *L. saxatillis* and *L. obtusata* using next generation sequencing methods. After *de-novo* assembly, 26696 putative transcripts were identified. These putative transcripts were compared with 1513 peptides identified through shotgun proteomic analyses applied to three male (testis, seminal vesicle and prostate) and two female (ovary and bursa) reproductive tissues (RTs) of *L. saxatilis*. This comparison identified 7 good candidates RPs that are being sequenced in all *Littorina* species.

SEQUENCING THE GENOME OF FRAXINUS EXCELSIOR (EUROPEAN ASH)

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Fraxinus excelsior (European Ash) is a common tree in Europe with about 80 million individuals in the UK, and is of great ecological and economic value. Ash is diploid, with 2C = 46 chromosomes. Ash trees in Europe are now threatened by the fungal pathogen *Hymenoscyphus pseudoalbidus*; it is estimated that around 99% of the ash trees in the UK are at risk of infection. Natural genetic variation within Ash populations includes a small percentage of low susceptibility individuals, as has been confirmed by a field study in Denmark. However, until now there is no reference sequence available which made it impossible to study the genetic variants that confer a low susceptibility phenotype on a genome-wide scale. We extracted DNA from a young ash tree produced by the self-pollination of a low heterozygosity tree from the Cotswolds in England. We measured its 1C genome size with flow cytometry as 880Mb. We are sequencing this *de novo* using a combination of Illumina HiSeq2000 and Roche 454 FLX+ technologies at Eurofins. We are assembling the reads with the CLC Genomics Workbench and other *de novo* assemblers for comparison and annotating it using MAKER. This work lays foundations for the study of genome-wide polymorphisms in European ash, and selection of genes for resistance to *Hymenoscyphus pseudoalbidus*.

STRATEGIC SPERM ALLOCATION IN RELATION TO MALE AGE IN THE LEK-MATING LESSER WAX MOTH ACHROIA GRISELLA

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Theory predicts that males are selected to allocate limited sperm reserves in a way that maximises their lifetime fertilization success. Fertilization opportunities may decrease with male age and there is some evidence that males increase sperm expenditure with age. However, it remains unclear whether transferring more sperm with older ages reflects strategic sperm allocation or simply a higher number of sperm available to older males. To specifically test for strategic sperm allocation, we analysed copulation behaviour and sperm expenditure in *Achroia grisella*, a short-lived, aphagous, lekking moth with an elaborate pre-copulatory mate choice system based on ultrasonic sexual ornaments. We staged a single experimental mating between virgin males of different ages (day of eclosion to 12 days old) and randomly assigned four-day-old virgin females to test for effects of male age on copulation behaviour and sperm expenditure, including the proportion of available sperm that is transferred during virgin matings. We find clear evidence for strategic sperm allocation in relation to male age and discuss its implications for the significance of post-copulatory sexual selection in one of the most prominent model species for pre-copulatory mate choice.

FUNCTIONAL IMPACT OF GENE LOSS IN GENOME EVOLUTION AND ANIMAL DIVERSITY: THE CHORDATE OIKOPLEURA DIOICA AS A CASE STUDY

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What is the impact of gene losses in the generation of biodiversity is a crucial question in Evolutionary Biology that still remains largely unknown. Until now, gene losses have been often neglected because the proof for gene loss is negative and difficult to detect, especially in complex genomes outside bacteria and yeast. The state-of-the-art sequencing technology offers us for the first time the exciting opportunity to perform exhaustive genome-wide analyses to detect unambiguous cases of gene losses in animals with complex genomes. Our group has started an ambitious project with two main aims. First, we will sequence full genomes of a several individuals from many populations and species of the larvacean class to gain novel insights into the dynamics of gene losses, to understand the correlation of gene loss with phenotypic traits, and to test for evolutionary gene dispensability (EGD), a new concept that we have developed in this project. Second, we will perform systematic knockdown experiments and RNA–seq analyses to provide functional evidence for the impact of gene loss, and test for adaptive and neutral events of gene loss in the context of genetic robustness and gene network connectivity. In this project, we use the urochordate Oikopleura dioica (and other related larvacean species) as case study, because the recent genome-sequencing project, in which we participated, revealed that this organism exhibits the smallest genome and the largest genomic plasticity of all metazoans known to date, including an extraordinary amount of gene losses. Preliminary results show the feasibility to identify actual genes that are in the process of being lost in different populations, which appear as potential candidates of losses that can be adaptive.

HISTORICAL CONTINGENCY AND PARALLEL PARAPATRIC DIVERGENCE BETWEEN LAKE AND STREAM STICKLEBACK PAIRS OF VARIABLE AGE

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When genetic constraints restrict phenotypic evolution, diversification is predicted to evolve along the so-called line of least resistance, the leading axis of the underlying G matrix. To address the importance of such constraints and their resolutions, empirical studies on parallel cases of phenotypic divergence of different age are valuable. However such studies are very rare. Here we study the parapatric evolution of six independently evolved lake and stream stickleback systems from Iceland and Switzerland, ranging in age from a few decades to several millennia. Using phenotypic data, we test to which degree parallel evolution occurs among independent lake-stream freshwater systems as well as during the marine-freshwater transition. We furthermore investigate in both cases how the underlying evolutionary trajectories diverge through time. We find that strong and consistent phenotypic divergence occurred independent of time for both the parapatric lake-stream systems and for the marine-freshwater transition. The extent of phenotypic divergence differs however between the two countries studied here. This indicates that historical contingency partially shapes the phenotypic outcome of divergent selection in lake-stream environments. Moreover, our results suggest that parapatric phenotypic divergence can evolve along a common evolutionary trajectory for some trait combinations, independently of their evolutionary age. The directionality of change in these traits may however differ due to historical contingency or environmental constraints. Other trait combinations differ rather between the two investigated countries but less within, suggesting that the underlying G matrix of each country could be differentially constrained. Thus phenotypic plasticity may play an important role during the colonization of novel habitats, where adaptive peak shifts may be readily achieved.

FEMALES BITE BACK: SEXUAL CONFLICT AND THE EVOLUTION OF VENOM PROTEINS IN THE REPRODUCTIVE TRACT OF FEMALE ANOLE LIZARDS

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Reproductive proteins evolve rapidly in many species, yet the ecological significance of these proteins remains largely unknown. In this study, we investigate reproductive proteins in *Anolis* lizards, a system in which the ecological basis for a fertilization bias has been established. When female *A. sagrei* mate with multiple males, they preferentially use the sperm from larger sires to produce sons. Field experiments reveal that this bias may be adaptive, based on patterns of offspring survival. Using Next Generation Sequencing, we investigate genes that are expressed in the reproductive tract of female anoles. Comparisons with *Drosophila*, the system in which female reproductive gene expression has been best studied, reveal broad similarities in the genetic response to mating across these distantly related taxa. Additionally, we investigated the molecular evolution of a group of serine proteases that are differentially expressed after mating in the reproductive tract of female anoles. Of these serine proteases, some appear closely related to snake venom proteins. Due to the deep origin of venom toxin in squamates and the hypothesized origin of *Drosophila* reproductive serine proteases from digestive proteases, our results suggest that *Anolis* reproductive serine proteases and venom serine proteases could share a common phylogenetic origin. This would imply that digestive enzymes have been involved in the evolution of cryptic aspects of female choice in multiple mating systems.

TRANSCRIPTOMICS REVEALS PATTERNING AND PIGMENTATION GENES SHAPING THE HELICONIUS MELPOMENE WING COLOUR VARIATION

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The wing colour patterns of the Heliconius butterflies are an excellent system to study the evolution of adaptive phenotypes in nature. Previous work has made important progress on the identification of both patterning and pigmentation candidate genes controlling wing colour phenotypes on these butterflies, however, a deep understanding of the genetic and developmental networks shaping Heliconius wing variation is still missing. Here, we have applied a transcriptomics approach (RNA-seq and expression arrays) on developing wings of two morphological and phenotypically different races of the butterfly Heliconius melpomene, in order to make a comprehensive identification of such genes. Across the genome, over 300 transcripts associated with colour pattern phenotype were identified, from which several associated to structural and pigmentation genes. Additionally, the tiling of three major colour loci, namely HmAc, HmBD and HmYb, pinpointed four genes within those genomic regions whose expression differed between phenotypic races, being therefore in association to wing colouration and likely play patterning roles controlling phenotypic "colour switches". The information here derived constitutes a step forward in the understanding of the genes and pathways controlling colour pattern evolution in Heliconius.

ONTOGENETIC GROWTH MODELS AND LIFE HISTORY THEORY

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Ontogenetic growth depends on a number of factors, among them food availability and temperature. For example, compared to ideal conditions, cold-blooded animals mature later and at smaller size when food is scarce, but later and at a larger size when growth is retarded by low temperatures (this is known as the Berrigan-Charnov puzzle). In this talk we introduce a food- and themperature-explicit mathematical model of ontogenetic growth which allows us to interpret this and similar phenomena simply as life-history decisions of the organism seeking to maximise its expected reproductive output. We conclude by discussing future applications of our model to resource ecology of host-parasite interactions.

COOPERATIVE NICHE CONSTRUCTION FACILITATES ZOONOSIS IN PATHOGENIC BACTERIA

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The majority of emergent human pathogens are zoonotic in origin. Understanding the factors underlying the evolution of pathogen host range is therefore of critical importance in protecting human health. Classical evolutionary theory predicts that the evolution of generalism in pathogens will be subject to trade-offs, and hence reduced within-host fitness compared to specialists, as pathogens evolve to tolerate multiple host environments. Here we show that rather than passively reacting to host environments, bacteria can use niche construction via cooperative secretions to achieve host generalism. We use an epidemiological framework to show that cooperative niche construction strategies can outcompete both specialists and classical generalists under a wide range of realistic conditions. We then use a phylogenetic comparative analysis of 191 bacterial pathogens to show that larger secretome sizes are associated with a greater probability of zoonosis, in agreement with our theoretical predictions. Our results suggest that cooperative behaviour is a key factor in the evolution of generalism in bacteria, and that monitoring programmes focusing on the horizontal transfer of secreted proteins could help identify future emerging human pathogens.

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