Poster abstracts

The 3rd Finnish Symposium for Molecular Ecology and Evolution

University of Jyväskylä, Finland
Poster 1: "Informed movement: how information shapes the co-evolutionary outcomes of range-expansion"

Presenter: Katja Rönkä

Range expansions are rapidly exposing species to new threats, particularly at northern latitudes. Predicting changes to these ecological communities however requires detailed knowledge of who is at the range front. This is because some phenotypes (and genotypes) are more likely to disperse than others, and neighbouring phenotypes (social environment) can influence selection directly or indirectly. Nevertheless, (1) how social environments change with range expansion and (2) how they influence species interactions remains largely unknown. We are answering these questions with the reed warbler which is a favourite host of brood parasitic cuckoos in the core of its range but is currently unparasitized at their range front in Southern Finland (cuckoos are present but favour other host species). We conduct behavioural experiments in the field to assay phenotypic variation, and utilize massive-parallel sequencing techniques (e.g. RAD) to characterize population structure, potential founder effects and the levels of past and present gene flow with parasitized populations towards the range core. We know that reed warblers have been breeding in Finland for about 100 years, but the origin of the population and their co-evolutionary history with cuckoos remains unknown. Range expansions provide an opportunity to test co-evolutionary theory if the history of the range front population is known, therefore we hope that molecular tools will provide us insight into this population’s past.

Poster 2: "Isolation and characterization of new Flavobacterium columnare strains and its bacteriophages from fish farms"

Presenter: Heidi Kunttu

Fish are an important source of protein, and aquaculture has been the fastest growing food production sector over the past 20 years. However, the intensification of fish production in farming systems expose fish to bacterial infections. Flavobacterium columnare is a Gram-negative bacterium that causes epidemical columnaris disease at fish farms during the warm water period. This pathogen causes significant financial losses in the industry if the disease is not treated with antibiotics as soon as the first symptoms appear. In order to develop a bacteriophage-based control of F. columnare, we isolated 117 F. columnare strains and 64 F. columnare phages from 10 different fish farms in Finland and Sweden. Genetic characterization of the bacterial isolates was made using 16S RFLP and ribosomal intergenic spacer analysis, and the virulence of 25 isolates was tested using a rainbow trout challenge model. Most F. columnare isolates could be assigned into four previously identified genetic groups A, C, E and G. Of these, bacterial isolates belonging to group C and E were the most virulent. Phage host range analysis against a collection of 229 bacterial isolates showed that most phages infect bacteria in a genotype-specific manner, and only few isolates had wider host ranges. Phages infecting the most virulent bacterial groups were isolated. The diversity, lytic capacity and combined host range of our phage collection demonstrates its potential to be used in phage therapy.
Poster 3: Effects of fungicide (fluazinam) on invasive Colorado Potato Beetles (*Leptinotarsa decemlineata*)

Presenter: Shahed Saifullah

Fluazinam (FLZ) is an active ingredient to control late blight in potato. FLZ affects the growth and mortality in fungi due to its role as an uncoupler of oxidative phosphorylation resulting in depleted ATP production by mitochondria. Uncoupling effect may also cause cell death by inducing reactive oxygen species. Studies have shown that ATP depletion and higher oxidative damages are coupled with abnormal (stimulated) respiratory and metabolic rate. These changes in oxidative status and respiration can be marked by the higher activity of genes related to oxidative stress and respiratory chain. However, their effects on nontarget organisms found in the agricultural fields are still unclear. To have a clear idea about the effects of FLZ on a nontarget species, i.e. Colorado Potato Beetles (CPB), we hypothesize that FLZ will induce 1) energy deficiency, 2) oxidative stress, 3) increase respiration and metabolic rate and 4) changes in genes associated with oxidative stress and respiratory chain. If the fungicide shows concentration dependent effects on ATP content, oxidative stress and metabolic or respiratory rate, we can then hypothesize that 5) higher concentrations of the fungicide may cause higher mortality in CPB. We want to test the effects of FLZ under four different field related concentrations, i.e. 0.66µg/ml, 0.25µg/ml, 0.125µg/ml and control. Any effects on their survival and physiology would give us important information about its mode of action in nontarget species.

Poster 4: "Micro-RIP: Functional analysis of uncultivated microbes using radioisotope probing"

Presenter: David Hopkins

Despite the wealth of big data (genomic, transcriptomic, metabolomic...) that now can be obtained from environmental samples, it has proven fiendishly difficult to reliably understand soil and water microbiomes beyond simple descriptions of their community diversity. Firstly, it is often difficult to directly link the anyone microbial type to its function/s, especially in samples from complex environments that contain large numbers of genera and often contain significant quantities of relic DNA from deceased organisms. Micro-RIP (Microbe Radiolotope Probing) is an ambitious project to address both these problems by re-purposing ion torrent next-generation sequencing chips to measure radioactivity. By doing this we can record from one sample both the genetic identity of a microbiome (via standard PGM sequencing) and then test the radio labelled status of those same DNA/RNA strands, essentially giving both identity and function of each genera found within a given microbiome. This tool would allow any classical radioisotope experiment designed to determine microbial function to do so on the genetic scale. This would allow us to test a broad range of ecosystem processes from nutrient flows to carbon dioxide fluxes for 100s to 1000s of distinct coexisting microbe genera in just one analysis. By doing so Micro-RIP could help lift the lid off the ecosystems function ‘black box’ that exists for many microbiotas.
**Poster 5: "The rates of evolution of grasshopper song (Orthoptera, Acrididae, Gomphocerinae)"

Presenter: Nikita Sevastianov

In grasshoppers of subfamily Gomphocerinae, song is the main component of reproductive isolation. Among singing Orthoptera, acoustic communication in Gomphocerinae is most developed in terms of complexity of stridulatory leg movements, the number of sound elements and mating strategies. The aim of the work was to compare rates of song evolution in grasshopper species with different mating strategies, which imply simple and complex courtship behaviour. We analyzed calling and courtship songs from 120 pairs of closely related grasshopper species, which were selected from molecular phylogenetic trees. For the calling song analysis, 31 characters that included both sound and stridulatory leg movement parameters were chosen. Each sister pair was categorized into data subsets based on geographical overlap (allopatric or sympatric) or mating strategy (simple or complex courtship), and we calculated evolutionary rates of calling and courtship song divergence for each subset. We showed an increased rate of calling song evolution in species with complex courtship behaviour, however, this difference between mating strategies was only found for sympatric species pairs. Evolutionary rates of courtship song were also higher in species with complex courtship behaviour, but this difference was independent on the geographical overlap between sister species. These results suggest the leading role of sexual selection in speciation of Gomphocerinae.

**Poster 6: Changing the genetic code through diet: protein restriction leads to reduced 18s rDNA copy numbers

Presenter: Joannes van Cann

Virtually all species have multiple copies of the gene complexes coding for ribosomal RNA (e.g. 18s), which is thought to be required for its high level of transcription. Exact number of copies vary between species and between populations, but can also differ between individuals and between generations. However, the drivers of these rapid changes in rRNA copy number are virtually unknown, despite their importance in aging and cancer development. Recently, diet was shown to induce predictable changes in rRNA copy numbers in Drosophila melanogaster. We performed an experiment in bank voles (Myodes glareolus) to investigate whether diet could have the same effect in mammals. We assessed whether (1) protein restriction induced changes in 18s rRNA copy number, (2) whether this change was inducible in adult and/or growing individuals and (3) whether copy numbers were related to body size. Our results show that exposure to protein restriction during the growing phase, but not the adult phase, reduced 18s rRNA copy numbers by 15%. rRNA copy number was also positively correlated to body size but only significantly to a skeletal measurement at birth, indicating that only prenatal structural growth is, at least in part, related to rRNA copy number.

**Poster 7: TBA

Presenter: Noora Poikela
Poster 8: "Exploring pleiotropic effects of yellow gene in *Drosophila montana*"

Presenter: Jesse Mänttäri

Body color plays an important role in animal’s fitness as it can be part of visual communication, temperature regulation and sexual selection. Some pigmentation genes have been found to effect more than one trait through different biochemical pathways and these pleiotropic effects can have an impact on both survival and reproduction of an animal. In insect, pigmentation can be shaped by both ecological pressures and sexual selection. In earlier studies, yellow gene, one of the main melanization genes of dopamine pathway, has been shown to have pleiotropic effects, for example, on *Drosophila melanogaster* male mating success and desiccation tolerance. We have used CRISPR/Cas9 gene editing technique to induce a mutation (deletion) in coding region of yellow gene of *Drosophila montana*, and investigated pleiotropic effects of this gene by comparing different phenotypes of *D. montana* wild type flies to yellow mutant flies. Since courtship song, produced by male fly, is an important part of *Drosophila* species courtship and mating success, I tested if yellow gene has an effect on different fly song parameters. Besides, I have also investigated yellow gene effect on temperature tolerance (critical temperature minimum and critical temperature maximum, since temperatures have many effects from physiological processes to behavior and species distribution in ectothermic species.

Poster 9: "Trypanosomatid Parasitic Protozoan Infections and Their Effects on *Drosophila lummei* Malt Fly"

Presenter: Päivikki Puolakka

Trypanosomatids are a diverse group of single cell eukaryotic parasites infecting humans, animals and plants with either simple (one host) or complex (two hosts) life cycles. Insect trypanosomatids are a broad group of parasites with simple life cycles and variable effects on their hosts. Temperature is one of the most important abiotic factor affecting insects life history, however, the effects of infection on host’s ability to respond to temperature differences is poorly known. New aspects of invertebrate immune system can be revealed by studying gut infecting protozoa, as earlier studies have focused mainly on responses to bacteria and fungi. This project explores the effect of trypanosomatid infection on cold tolerance, survival and locomotor activity of *Drosophila lummei*, a northern highly cold tolerant fly species. Severity of the infections was measured using quantitative PCR (qPCR) and effects of infection on fly phenotype was tested with locomotor activity and survival tests. qPCR tests showed females to be less infected than males, and overall infection load to be lower in cold acclimated (+6C) flies when compared to non-acclimated (+19C) flies. Locomotor activity tests indicate that there is a slight difference in total activity between infected and non-infected flies in specific light-dark cycle and survival test showed that infected flies had a shorter life span. In conclusion, trypanosomatids seem to effect in several ways to *D. lummei* fitness traits.
Poster 10: "A gut feeling for development?"

Presenter: Anna-Lotta Hiillos

The marine polychaete worm *Pygospio elegans* can produce distinct kinds of larvae (poecilogony): planktonic larvae, capable of dispersal, and benthic larvae that do not disperse, but develop in brood capsules maintained by their mother. It is unclear what causes this polymorphism as neither a genetic nor an environmental basis for it has been determined. Parasitic marine gregarines in the genus Selenidium (Apicomplexa) have recently been found in the intestine of *P. elegans*. It is known that intestinal parasites can affect their host's biology in a variety of ways but the role of parasites on developmental mode evolution has not been explored. Our aim is to investigate if Selenidium sp. infection has an effect on physiology and larval developmental mode variation in *P. elegans*. We have performed a ddPCR survey using apicomplexan Cox 1 – gene to determine the intensity of parasite infection in four *P. elegans* populations with different main modes of larval development in Isefjord-Roskilde fjord estuary (Denmark) with seasonal sampling of 4 times within a year. Preliminary results suggest that Selenidium sp. is most abundant with benthic larval mode throughout the year and the infection load increases towards late fall in all populations.

Poster 11: "The cases of introgression in evolution of the Drosophila virilis species group (Diptera: Drosophilidae)"

Presenter: Svetlana Sorokina

The introgression of genome parts often becomes a cause of erroneous phylogenetic conclusions. Incomplete reproductive isolation, asymmetric hybridization and population size fluctuations may favor distribution of obtained genetic material. The drosophila virilis group is the group of closely related species, some of which are on the early stage of divergence. We compared three sets of phylogenetic data: the trees based on the mitochondrial cytochrome c oxidase subunit I (Cox1), on the Y-chromosomal 1-beta dynein heavy chain (kl-2) gene sequence and on nuclear autosomal markers. We have identified two cases of contraventions of tree topology, which may be caused by the introgression. The first case is associated with *D. lacicola*, a North American species closely related to *D. montana*. The data of nuclear markers revealed the differentiation between these taxa, which was supported by the data of Y-chromosomal marker. However, the mt-haplotypes of *D. lacicola* are placed into spectrum of mtDNA diversity of North American *D. montana*, that makes *D. lacicola* and *D. montana* paraphyletic species. In the second case, we investigated *D. borealis*, a North American species diverged from montana phylad in the beginning of montana subgroup evolution. According to mitochondrial and nuclear genome data, its position on the tree is different in relation to *D. flavomontana*. The Y-chromosome marker data supported the mitochondrial tree but not the tree built on the base of autosomal markers.
**Poster 12: Multi-gene expression patterns during anhydrobiosis in *Hypsibius exemplaris***

Presenter: Tiia Nikupaavola

Anhydrobiosis is a phenotypic adaptation common among microinvertebrates, like tardigrades, representing survival strategy to periods of severe dehydration. Gene expression analyses provide a window to the molecular mechanisms enabling survival and recovery from desiccation. Proteins of three major groups have been suggested to have a key role in such adaptations: heat-shock proteins (HSPs), aquaporins (AQPs) and late embryogenic abundant proteins (LEAs). Unlike most studies before, our study combine the simultaneous estimate of expression levels in all three key protein groups and a comparison across consecutive anhydrobiosis states using the tardigrade *Hypsibius exemplaris*. By using digital droplet PCR (ddPCR) we could obtain more precise expression measures. Our results show different patterns of gene expression in all target genes: HSP70-like 1 levels halved from the active state to the preconditioning state and dropped by 80% in the dry state compared to the transition state, but levels between active and transition states stayed similar. AQP10 levels had 3 times more in both active and dry states and 5 times more in the precondition state than in the transition state. Expression of LEA1 showed induction during the preconditioning state. The findings from HSP70 resemble expression changes in HSP70-3 found previously in the tardigrade *Milnesium tardigradum*. AQP and LEA induction during precondition suggest these genes are crucial for *H. exemplaris* to enter anhydrobiosis.

**Poster 13: "DNA methylation and gene expression patterns in a fish meta-population subjected to variable thermal conditions"**

Presenter: Tiina Sävilammi

Understanding epigenetic landscapes and how epigenetic variation affects gene expression is an intriguing question that can be addressed with the help of whole-genome NGS-methodologies. Here, we study a meta-population of European grayling (*Thymallus thymallus*), a spring-spawning, freshwater salmonid fish with high homing propensity that has undergone contemporary evolution of early life-history traits in response to temperature despite low genetic variation. We performed a common garden experiment in which embryos originating from 5 subpopulations and 2 thermal origins were raised in temperatures resembling the alternative natal thermal environments, and used bisulfite sequencing of 30 hatching stage embryos to conduct a low coverage genome scan for methylation percentages of methylation sites. We combined these data with RNAseq data from the same populations and a chromosomal level genome assembly. We found that methylation levels are affected by the location in the chromosome (intergenic, regulatory and protein coding regions) and validated that methylation, particularly in the regulatory regions, is associated with lower gene expression. We found a global pattern of methylation downregulation (gene expression upregulation) in the cold rearing environment and more specific candidate genes that might be important for thermal plasticity.