2020

KAAMOS

Symposium

Abstract Booklet

24th Kaamos Symposium

9th – 11th December 2020

University of Oulu

FACULTY OF SCIENCE
UNIVERSITY OF OULU

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Temporal genomics as tool for applied conservation

Prof. Tom Gilbert

Exactly what happens to species' genomes and genetic diversity as they go through massive contractions is a question that has held the fascination of biologists for decades. This question is not just of theoretical interest, but critical to developing strategies to help preserve our increasingly threatened biodiversity in which population collapses globally as a result of anthropogenic actions are becoming increasingly common. Although theory can be used to provide some insights, and laboratory experiments others, ultimately biology is sufficiently complex that the only way to truly understand this problem is through real datasets. Ideally these require population scale sampling of not just the present, but the past, in particular from specimens pre-dating the most recent collapses. Population palaeogenomic tools are becoming an increasingly powerful and cost-effective approach for generating such data. In this talk I present some examples of how such approaches can work, and what they reveal.

The legacy of population bottlenecks: Studying how the woolly mammoth became extinct and muskox is living on the edge

Patrícia Chrzanová Pečnerová

Evolutionary history of many species is shaped by past population bottlenecks that tend to lead to reductions in genetic diversity. However, while some species seem to be negatively impacted by these reductions, others persist and even show increasing population trends. Therefore, we analyzed genomic data from the last population of the woolly mammoth and from Greenlandic muskoxen, two species representing the two sides of the spectrum. Mammoths and muskoxen share many traits of a typical Ice Age megaherbivore, and our results suggest that both species suffered genomic consequences of extreme founder events. Studying why one species became extinct, while the other species survives, can aid our understanding of the microevolutionary processes that take place in populations living on the brink of extinction.

Patterns of genomic structure in parapatry: a case study of *Melitaea athalia-Melitaea celadussa*

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The parapatric mode of distribution involves species with mostly non-sympatric ranges but with a narrow area of overlap. This pattern creates an interesting yet challenging system for defining species boundaries. As gene flow between closely related parapatric taxa is known to take place frequently, we aimed to unravel the patterns of genetic structure in a parapatric butterfly system. The ranges of our model species cover a vast area of Europe and the western parts of Asia and exhibit a narrow contact zone in France, Switzerland, Italy and Austria, with *Melitaea celadussa* occurring south and west and *M. athalia* north and east of it. We used high throughput ddRAD sequencing method to recover a large number of informative loci. Most *M. athalia* showed a noticeable phylogenetic paraphyly with regards to *M. celadussa*, suggesting extensive introgression. Remarkably, our phylogenetic tree revealed a well-separated clade of *M. athalia* from the Balkans. A demographic population history analysis indicated that the paraphyletic *M. athalia* represents an intermediate group between *M. celadussa* and the Balkan clade. Analyses of genomic structure revealed a significant amount of admixture between the two taxa especially within the continental European *M. athalia*. We conclude that there has been historical, and possibly ongoing, gene exchange between the two parapatric taxa. This phenomenon may theoretically result in speciation or merging of the two species in the future. The admixed individuals seem to have gained a capacity to spread northward. This could have been enhanced by a selective advantage for admixed specimens, possibly through traits that improve the survival in areas with cooler climate.

Keywords: Parapatric, paraphyly, contact zone, admixture, gene exchange

Leptidea butterflies as a model for research on speciation and chromosomal evolution

Vlad Dincă

Leptidea Wood White butterflies represent one of the most remarkable examples of cryptic species in Eurasian butterflies. In particular, *Leptidea sinapis* has been intensively studied during the last two decades, resulting in the discovery of two cryptic species (*L. reali* and *L. juvernica*), which have been detected based on genetic differences and reproductive isolation due to female mate choice.

Infection with the maternally inherited bacterium *Wolbachia* is also widespread in *Leptidea*, and patterns of infection suggest that horizontal transmissions may have taken place. Laboratory experiments on different populations of *L. juvernica* also indicate the presence of *Wolbachia*-induced cytoplasmic incompatibility.

This butterfly genus also displays a notable karyotype variability, which is particularly pronounced in *L. sinapis*. As a matter of fact, this species is currently the metazoan with the highest intraspecific chromosome number variability (2n=56 to 2n=106), not related to polyploidy. Laboratory mating experiments of geographically distant chromosomal races (separated by at least 24 chromosomal fusions/fissions) have shown that hybrids displayed an unexpectedly high reproductive fitness (42% of that of the control lines). This tolerance to chromosomal rearrangements may be facilitated by inverted meiosis.

The recently acquired knowledge and ongoing discoveries related to *Leptidea* butterflies make this genus an emerging model for the study of speciation and chromosomal evolution in organisms with holocentric chromosomes.

Female emancipation (from care): how, why & when?

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Conventional sex roles imply that females look after the young whereas males contribute little to parental care and rather seek additional matings with other females. These sex roles are widespread across mammals and birds. In some species, however, females have managed to emancipate themselves from this stereotypic role. In Snowy Plovers Charadrius nivosus, parents incubate jointly the eggs but soon after hatching of the chicks, many females leave their families and become polyandrous by starting a new breeding attempt with another male. Their deserted partner will then diligently look after the chicks alone. But not all Snowy Plover females follow this mating strategy: some females opt to stay and care for their offspring until they are independent. Using data collected from a population of Snowy Plovers breeding at Bahía de Ceuta, Mexico, I will show that individual females are extremely flexible in their care and mating behaviour. Female polyandry has no relationship with age but is associated with egg size: polyandrous females produce smaller eggs than monogamous females. In our study population, polyandry is helped by a male biased adult sex ratio that is driven by better survival of males throughout all life stages with particularly high differences during the first year of life. When females desert their broods, they take into account the value and prospect of their present brood whilst considering future reproductive opportunities. This decision-making process is highly flexible and dynamic demonstrated by a close temporal co-occurrence between female desertion and chick mortality. These results provide deep insights into the process of female emancipation from parental care. Females are sensitive to multiple social cues when deciding whether to care or desert their offspring.

Extended phenotype as a signal in interspecific information use

Reetta Hämäläinen

The extended phenotype entails both the organism's physical body and alterations it induces to its immediate surroundings. Nests are a typical example of extended phenotypes and studies have shown that bird nests are an important source of information in an interspecific context. Pied flycatchers use the nests of great tits as a signal for a good breeding site and they gain fitness benefits by breeding close to their potential competitor. Flycatchers also use the visible clutch size of tits as information of how much to invest in their own offspring. However, it is not known how great tits' phenotype and visible clutch size affects their settlement decision and what are the full costs and benefits of coexistence for flycatchers. Our aim was to examine how a physical phenotype (tarsus length) of great tits and flycatcher and the clutch size of a great tit explains the nesting site decision of flycatcher to breed either near or farther away from tit nest and whether there is a fitness consequence for the flycatcher. Our results show that great tits clutch size did not have an effect on the pied flycatcher settlement decision. However, the great tits physical phenotype had an effect. Flycatcher females were more likely to nest near great tits when there was a bigger great tit female present at the site. Additionally, flycatcher female's investment to their offspring (egg mass) was greater when there was a bigger great tit female present, regardless of the settlement decision.

Complexities of small passerines’ diets

Coen Westerduin

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Knowledge on the connections in food webs is fundamental to our understanding of ecosystems. In addition, such information has significant implications for our understanding of evolution and biodiversity, of communicable diseases or bioaccumulation of toxins, while also yielding applications in agriculture, forestry, conservation and beyond. Because of this importance, numerous methods have been used in the past few decades to unravel the highly complex networks of interacting species that occur in nature. Only with recent advances in DNA-based methods, however, has it become possible to identify the species involved in some of these interactions with the accuracy, detail and ease required for large-scale applications. Here, I will present results from ongoing DNA metabarcoding research on the diets of several local passerines. Faecal samples from the birds’ chicks were collected during the most energy-demanding phase of the breeding period in spring, a period which several of these birds time to correspond with a seasonal peak in food availability. As the birds are insectivores, thousands of different species could potentially be part of their diets, and in our study, their diets have already been found to encompass many hundreds of these. While broadly alike at the order level, more detailed species-level investigations reveal highly variable patterns of prey overlap and differentiation for the diets of these seemingly similar birds. These patterns will be explored using several metrics and conducted using various taxonomical and categorical groupings to elucidate the exact differences between these passerines, and how they interact with their environment. In turn, this information can help us understand how the largely anthropogenic changes to these environments — climatic change that is shifting the seasonal timings, and forestry which is already seen as a major driver in the population declines observed for some of these birds — may impact forest ecosystems in the decades to come.

Novel species interaction indices complement biodiversity measures

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The importance of biotic interactions in biodiversity measures has been acknowledged. We aim to supplement the current practices of measuring biodiversity, taxonomic (TD), functional (FD) and phylogenetic (PD) diversity, by introducing two novel interaction indices, Characteristic interaction index (Ichar) and Interaction dispersion index (Idis). These indices will define the sign and strength (Ichar), and the diversity (Idis) of species interactions within a community. The indices will supplement the current shortcomings in estimating the diversity of biotic interactions as they can widely be applied to (i) a variety of taxa, (ii) within or across trophic levels, (iii) spatial, temporal or spatio-temporal data, and (iv) spatially extensive data.

The indices are derived from species-to-species covariance matrices produced by Joint Species Distribution Models (JSDMs). In this study, first, we used a simulation experiment to analyze the behavior of the indices in relation to community structure. Second, to demonstrate the use of the indices, we derived them for natural breeding bird communities by using a dynamic JSDM. We used large spatio-temporal datasets of forest and farmland birds in Finland and six biogeographic regions in France. Finally, we compared the Interaction Dispersion index (Idis) to indices of TD, FD and PD for the natural bird communities.

The simulation experiment showed that Ichar and Idis were robust to variations in abundance distribution of species and assumptions concerning the accuracy of the species-to-species covariance matrix. Thus, Ichar and Idis appear able to capture biological variation in species interactions among communities without being confounded by community structure or methodological accuracy. All bird communities had a positive Ichar and farmland communities had stronger positive interactions than forest communities, except in Finland and Alpine region. Idis was also higher for farmland communities, excluding Finland. Mediterranean farmland region had the highest Ichar and Idis. Idis tended to have a positive correlation with each of TD, FD and PD, yet only correlation with FD was statistically significant. Ichar and Idis were strongly correlated ($r = 0.88$) in bird communities.

The importance of including biotic interactions in biodiversity estimation is known, yet the methodology for this has been lacking. Current statistical methods enable the estimation of species-to-species abundance covariances from large datasets, facilitating the derivation of novel interaction indices to supplement biodiversity measures. Ichar and Idis provide a tool for quantification of the diversity of species interactions, which facilitates new insight into biodiversity variation among communities.

An insight into host-endosymbiont interaction in *Pinus sylvestris* L.

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Intracellular and colonization patterns exhibited by Methylobacterium extorquens DSM 13060, the facultative methylotrophic endosymbiont of Scots pine (*Pinus sylvestris* L.), shows their ability to utilize methanol as source of carbon and energy. Previous evidence shows that methanol is released by the phyllosphere of plants as a by-product of pectin metabolism in biosynthesis of cell wall. These metabolic resources facilitates microbial colonization in plants niche. Data generated from the genomic analysis reveals the importance of genes associated with endophytic lifestyle and processes, such as methylotrophy, where they are clustered in the form of an operon. The gene mxaF is known to be the core, responsible for controlling the transcriptional processes for methanol oxidation. Another important process included is biosynthesis as well as degradation of poly-β-hydroxybutyrate (PHB). M. extorquens DSM 13060 like other members of the Methylobacterium genus accumulates PHB intracellularly as carbon reservoirs in both single and multi-carbon substrates. Major genes involved in the biosynthesis and degradation of PHB includes phaA, phaB, phaC and phaZ which encodes β-ketothiolase, NADPH-linked acetoacetyl coenzyme A (acetyl-CoA) reductase, PHB synthase and PHB depolymerases, respectively. However, another gene phaR, reportedly plays a key role in the acetyl-CoA flux to PHB indicating its major part in the activity of the serine cycle.

During progressive colonization, M. extorquens DSM 13060 indicated high-level expressions of promoter-reporter mxaF, phaC and phaZ (phaZ1-Z2) on epidermis, cortical cells and parenchymatous cells throughout root tissues and the transition zone between root and stems. This signifies an ongoing methanol assimilation and storage in accordance to several earlier reports where mxaF promoter is highly active in epiphytic or nodule-bound lifestyles. Promoters of phaZ1, phaC and mxaF were activated during infection of outermost tissues of pine seedlings, in the cylindrical sheath, epidermal layer and outer cortex. Hence, the role of mxaF, phaA, phaC and phaR in the process of carbon cycling and storage are studied with mutants deficient of the genes. This has been achieved with generating a platform, where the bacteria harboring the mutants are tagged with green fluorescent pigment (gfp) and the wild type with red fluorophore, ruby (mRuby2), co-cultured to attain a comparative platform and subsequently tracking them in the host system.

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Testing the masking hypothesis in predominantly diploid conifer *Pinus Sylvestris*

Sandra Cervantes

There is a wide variation on ploidy level among living organisms. However, the evolutionary mechanisms underlying these differences are still not well understood. One of the most studied ploidy systems is the alternation of haploid and diploid life phases. Theory predicts that haploid and diploid stages will be affected differently by selection. Genes expressed in haploid stage will respond faster to selection, as deleterious mutations will be immediately expressed and beneficial mutation will reach fixation faster. In contrast, selection will be less efficient in genes expressed in diploid stage, where the fitness effects of recessive deleterious or beneficial mutations can be hidden from selection in heterozygous form. Theory expectations have been confirmed in yeast, however it is not completely clear if these hold for more complex organisms. Results from studies of evolutionary rates of genes expressed in haploid or diploid stage in *Arabidopsis thaliana*, *Capsella grandiflora*, and the moss *Funaria hygrometrica* are inconclusive. Here we propose to use *Pinus sylvestris* to identify genes expressed in haploid (megagamethophyte) and diploid tissues (embryo, bud, needle, and phloem). We generated 30 transcriptomes (five tissues with six biological replicates per tissue), and we have used differential expression analysis (DE) and the tau index to find tissue-specific expressed genes in our samples. Next, by using targeted resequencing, we plan to analyze the genetic diversity and allele frequency spectra of some of the genes expressed in haploid and diploid stages to study the effect of selection on them. We expect our results to contribute to the understanding of evolutionary dynamics affecting different ploidy levels.

Composition of endophytic communities in wild berry fruits is shaped by host plant species

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Wild berries are interesting research subjects due to their rich sources of health-beneficial phenolic compounds (Manganaris et al., 2014; Zafra-Stone et al., 2007); however, the microbial communities residing in wild berries (endophytes) are rarely studied. Endophytes are bacteria or fungi which live inside the plant tissues, and their functions vary depending on the host plant species and environmental parameters (Hardoim et al., 2015). The present study aims to examine the community composition of culturable endophytes in fruits of three wild berry species (bilberry Vaccinium myrtillus L., lingonberry Vaccinium vitis-idaea L., and crowberry Empetrum nigrum L.) and investigate the effects of host plant species and growth sites in shaping the endophytic communities.

We applied the culture-dependent approach to study the endophytic microbes in the wild berry fruits. We performed PERMANOVA test on the Raup-Crick dissimilar matrix of the endophyte communities to examine the effect of host plant species and growth sites on the endophytic community assembly.

Two fungal phyla (Ascomycota and Basidiomycota) and one bacterial phylum (Proteobacteria) were detected altogether. We identified previously unknown endophytic fungal taxa in the wild berry fruits including Angustimassarina, Dothidea, Fellozyma, Pseudohyphozyma, Hannaella coprosmae, and Oberwinklerozyma straminea. The composition of endophytic communities was different among the berry species (PERMANOVA, $P = 0.01$), while it did not differ among the growth sites (PERMANOVA, $P > 0.9$).

We conclude that host plant species had a strong effect on shaping the endophyte communities in the studied wild berry fruits. Moreover, wild berry fruits are reservoirs of diverse endophytic taxa which can be exploited for their benefits.

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Composition, biosynthesis and effect of environmental factors on cuticular wax of bilberry

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Keywords*:* Bilberry, lingonberry, berry press cakes, cuticular wax, triterpenoids, glossy mutants

The outer surface of plants is covered by cuticular wax, which plays a role in non-stomatal water loss, protection from UV rays and plant defense. We have studied the chemical composition of cuticular wax in bilberry (*Vaccinium myrtillus*), lingonberry (*Vaccinium vitis-idaea*), bog bilberry (*Vaccinium uliginosum*) and crowberry (*Empetrum nigrum*) fruits using GC-MS. Triterpenoids, known for their health beneficial properties were found to be dominant compounds in bilberry and lingonberry cuticular wax. Therefore, the industrial leftover of berry (bilberry and lingonberry) juice industry, berry press cakes were utilized to extract wax using clean and environmentally friendly technique, Supercritical Fluid Extraction (SFE). Linoleic acid and γ-linolenic acid were found to be the dominant compounds. Berry waxes show good *in vitro* Sun Protection Factors (SPFs) depicting high UV-B absorbing capacities. Developmental and environmental factors play an important role in cuticular wax biosynthesis. Therefore, in the next study glossy wax mutants and wildtype bilberry were studied through the course of fruit development. The wax load between mutant and wildtype bilberry was found to be almost similar, however the proportion of triterpenoids was higher; fatty acids, aldehydes and ketones, lower in mutant wax as compared to wildtype bilberry during development. Peel specific expression of wax biosynthetic genes such as *CER26-like*, *FAR2*, *CER3-like*, *LTP*, *MIXTA*, and *BAS* indicates their role in wax biosynthesis in bilberry. While studying the effect of environmental factors on bilberry cuticular wax, we observed that the proportion of triterpenoids increases in as we move from northern latitudes to southern, and correlation analysis suggested temperature to be a major influence. Controlled chamber phytotron study confirmed the role of temperature in increasing the proportion of triterpenoids in bilberry cuticular wax. Our study brings new information on composition, biosynthesis and effect of environmental factors on bilberry cuticular wax.

A role of hybridisation for biodiversity?

Prof. Jochen B. W. Wolf

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Genome-wide data accumulated over the last decade has clearly demonstrated that hybridization plays an integral role in the evolutionary history of many species. Hybridization may slow down or reverse species divergence, accelerate it via adaptive introgression, induce instantaneous isolation by allopolyploidization or open novel niche space to transgressive genotypic combinations. In this seminar, I will address the role of hybridization from two perspectives in the same system, the fission yeast *Schizosaccharomyces pombe*. The first perspective considers the role of gene flow on adaptive divergence which we explored in an evolution experiment (Tusso et al. 2020). The second perspective considers the consequences of ancestral hybridization in natural populations of fission yeast (Tusso et al. 2019). These examples illustrate the importance of migration with subsequent hybridization on the emergence and maintenance of biodiversity.

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Impacts of gold mine effluent on water quality in a pristine sub-Arctic river

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Impacts of mining on water quality are a great concern in the Arctic region. This study evaluated the impact of pre-treated mine effluent on river water quality. The study was conducted along the Seurujoki River in sub-Arctic Finland, which is impacted by Kittilä gold mine. The study analyzed water quality and hydrological data upstream and downstream of the mining area over an eight-year period, including a tailing dam leakage event in 2015. The analysis focused on water quality determinants such as electrical conductivity (EC), sulfate, antimony, manganese, and total nitrogen (N_{total}). Descriptive statistics on river water at four stations along the river corridor showed negative impacts of mining activities on the recipient water body. In order to find an indicator for water quality, correlation analysis between the water quality determinants was carried out. It identified EC as a good indicator for continuous water quality monitoring, especially to detect mining accidents such as partial failure of a tailings dam. The results showed increasing contaminant concentrations due to mining as more mine effluent was generated over time. A linear mixed model was developed to predict the coefficient of different elements affecting EC at river water monitoring stations impacted by mining effluents. The results provide new information on how to assess mining water impacts and plan future water quality monitoring.

Keywords: Mining, environmental impacts, contamination, accident, Finland.

Usability of microbial communities as environmental indicators in mine area and as tools for biological remediation in an open mine pit lake

Tiina Laamanen

Mine closure is an essential part of the development of sustainable mining industry. In Finland, we are in a unique situation as in the near in the future several mine sites are being closed according to current legislative requirements. In the ERDF-funded Mine closure -impacts and possibilities -project we are addressing the new potential of closure facing mine sites. Project partners are the University of Oulu Kerttu Saalasti Institute, the Geological Survey of Finland (GTK) and Finnish Environment institute (SYKE). The study site of the project is the former Hitura nickel mine, located in Nivala, Northern Ostrobothnia.

SYKE is responsible for the implementation of work package 3 “Use of an open mine pit lake as a bioreactor to remove metals and sulfate”. The aim of the work package is: 1) Investigate the suitability of microbes for cleaning in laboratory-scale bottle experiments using wood chips, biogas plant reject and pig manure, 2) Test further these materials and conditions in a larger mesocosm scale container test simulating the conditions in an open mine pit lake and 3) Create guidelines for removing metals and sulfate from open mine pit lake’s water.

We collected water to our laboratory-scale bottle experiments from Hitura open mine pit lake. We took waters from a depth of one meter, 45 meters 88 meters. In the laboratory, wood chips, biogas plant reject, and pig manure was added to 1 liter of water from three different depths. All the test was performed as a triplicate. Eight weeks incubations were performed at +10 ‘C Preliminary results from laboratory-scale bottle experiments show that using the pig manure works the safest - in all cases the results were good. We found that biogas plant reject used in our tests contains some heavy metals, so it is not a good option for that kind of purpose. Wood chips are a good breeding ground for bacteria, but slow decomposition maybe inhibits the effectiveness.

However, more detailed conclusions are still needed after the analysis and interpretation of results for the water samples of larger mesocosm test and DNA sequencing of microbial samples taken from both bottle-scale and larger mesocosm-scale tests.

Flying microbes – first results from the EOSTRE stratospheric balloon flight experiment

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Earth's stratosphere (~10 to 50 km altitude) is characterized by hypobaric conditions, low temperatures, high intensities of UV and cosmic radiation as well as low water and nutrient availability. Even though it is not considered a permanent habitat for microorganisms, these can be transported to the stratosphere by vertical air movement caused by storms, volcanic action or human activity (e.g. weather balloons, aircrafts, rockets or satellite transfer).

The EOSTRE project (Experiment on Outliving microorganisms under STRatospheric Environment) aimed to test the influence of the extreme conditions in the stratosphere on microorganisms and their survival as well as on possible mutagenic effects of the strong radiation. A sample gondola was built with inclined sample holders that allowed exposure of endospores and known extremophilic microorganisms to radiation at different intensities. Moreover, three environmental samples (soil and sediment) were placed in the sample gondola to test the effect of stratospheric conditions on complex microbial communities.

The sample gondola was attached to a stratospheric balloon which was launched near Kiruna, Sweden, on November 3rd, 2020. The balloon ascended to a height of approximately 25 km and left to drift eastwards for 1.5 h (covering a distance of approximately 200 km) after which the balloon was capped to initiate descent near Pokka, Finland. During the flight, the samples was exposed to pressures as low as 2 kPa and temperatures as low as -50ºC as well as high UV radiation. First experiments have been started with the environmental samples as well as with the pure cultures.

For the environmental samples, the effect of stratospheric conditions was assessed by Most Probable Number (MPN) counts of aerobic and anaerobic heterotrophs and arsenic-tolerating microorganisms as well as by substrate-utilization tests. Based on the first results, the microbial communities in the three environmental samples reacted differently to the stratospheric conditions. Cultivation was possible from all three samples, indicating that at least part of the microbial community had survived. Compared to controls, for two of the samples that had been to the stratosphere MPN counts of approximately one to two orders of magnitude lower were obtained, while similar MPN counts were obtained for the third sample. This indicates that the survival rate of culturable microorganisms was at least ~1% for all tested environmental samples. This rather high survival rate might have been caused by the sample matrix which helped to protect the microorganisms from radiation and desiccation during the flight to a certain degree.

Owls for Peace

Prof. Alexandre Roulin

The barn owl is a fascinating bird that I am studying for more than 25 years. It is cosmopolitan, its plumage varies in coloration and this bird shows remarkable behaviour including cooperation between young siblings. In this talk I will highlight some key results our research group obtained on the study of

(1) colour polymorphism: barn owls vary in the degree of melanin-based coloration from white to reddish and from spotless to heavily marked with black spots. The degree of reddish coloration is important in the context of natural selection (predator-prey interaction), whereas the size of black feather spots is important in the context of sexual selection (male mate choice).

(2) reproductive behaviour: barn owls have interesting sexual behavior such as extra-pair copulation and divorce.

(3) social interactions between family members: The barn owl is exceptionally peaceful. Nestlings preen each other, they feed each other and they vocally negotiate which individual will have priority access to food instead of fighting.

The barn owl is thus an inspiration for us as human. We thus use this bird to bring Israeli, Jordanian and Palestinians at the same table. Farmers spread poison to kill rodents that devastate their agricultural fields. This is an ecological disaster because predators that consume poisoned rodents accumulate poison and die. We thus motivate farmers to use barn owl as environmentally-friendly biological pest control agent. To implement this project in the t hree regions we organize workshops with Israeli, Jordanian and Palestinians. This approach is of interest to diplomats, farmers, state leaders and even the Pope received us to explain the relevance of our ecological project in reconciliation between communities in war.

Information of wolves’ movements decreases wolf attacks on dogs

Mari Tikkunen

Wolf *(Canis lupus)* attacks on hunting dogs are a major conflict between wolves and hunters in Northern Europe and Northern America. Also, in Finland, the threat that wolves pose to hunting dogs is one reason why Finnish hunters have negative attitudes towards wolves and one of the potential motivations for the illegal killing of wolves.

During 2010–2017, wolves killed an average of 38 dogs (range 24–50) per year in Finland. Most of the attacks (91%) were directed at hunting dogs during the hunting season. The reasons for attacks are poorly known, and there are only few solutions to prevent the attacks.

We studied if knowing where the GPS-collared wolves are decreases the risk of attacks by comparing the distribution of territories of collared and uncollared wolves, with and without attacks.

In Finland the last positions of GPS-collared wolves were accessible to the public with a 5 x 5 km resolution during hunting seasons of 2013–2014 and 2015–2018. The link was visited more than 1 million times in 3 of the 4 seasons. Fatal attacks on dogs occurred on 17% of the days during the hunting seasons of our study (n = 760 days). Both the attacks and visits peaked in September–November, which is the primary hunting season in Finland. The number of daily visits to the website was higher on days when fatal attacks occurred than on other days.

The proportion of attacks that took place in the territories without any GPS-collared wolves was higher (50%, n = 48) than that in the territories with at least one collared wolf (32%, n = 22, chi-square for difference = 7.86; p = 0.005; n = 48).

Although public information decreased the risk of attacks, it did not completely protect dogs from wolf attacks and may in some cases increase the risk of illegally killing wolves. The most remarkable benefit of this kind of service to the conservation of the wolf population might be the message to the public that management is not overlooking hunters’ concerns about wolf attacks on their dogs.

Using new bio-logging technology for tracking wildlife animals and their behaviour description

Václav Silovský

The bio-logging technology is mostly used for small animals like avian or marine species. Devices usually allow high frequency tracking of individuals in short term schedule which is caused by minor batteries. Bio-loggers are developed for recording accelerometer and magnetometer data on defined frequency and provide detailed information about animal's body acceleration and magnetometer orientation. In our study we have developed new technology based on using biologgers in combination with traditional and widely used GPS collars appropriate for long term monitoring. This new hybrid technology allows observation of large animals for long-term studies and provide detailed datasets of their behaviour and movements. The hybrid collars come up with the data in different ways: movement reconstruction, orientation and animal behaviour in a very fine scale which was impossible in the past.

Keywords: Bio-logging technology, animal behaviour, GPS telemetry, movement ecology

Behavioral reaction of wild boars to human activities in the forest and its vicinity evaluated by using data obtained from biologgers located in animal's collars

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Wild animals may perceive human activities as a threat. As a result of the threat, animals change their spatial orientation and distribution. Based on the risk of threat animals should avoid human presence and react as they should during a conflict with a predator. Regarding to the fact that the previous studies of the effect of disturbances in large mammals were limited by technological progresses, we decided to use modern biologging technology to describe in detail the reactions of animals, which has not yet been possible in wild animals. We investigated the effect of human presence on the spatial orientation of wild boars (Sus Scrofa) in their natural environment. For the analysis, we selected a total of 4 types of human activity – driving a car, a walking tourist with and without a dog and cycling. Using hand-held GPS, we recorded the positions of selected types of human presence. The localities in which we monitored in 2019-2020 are Doupov and Kostelec nad Černými lesy, Czech Republic. We obtained data from biologgers located in animal's collars (collars contained GPS, 3-axis high resolution accelerometer and magnetometer). Subsequently, we processed the data in DDMT software using dead-reckoning, which is a unique tool for describing the movements of animals on a fine scale. The method allows to reconstruct the exact track which was impossible in the past. We evaluated the results in RStudio and used QGIS for data visualization. Wild boars mostly avoided human dwelling, but did not avoid or change the spatial distribution in the forest, where was the human disturbance. The study of animals behavioral and movement responses is important because it can help take action against spreading of African swine fever and define the measures in the different environment.

Keywords: wild boar, human disturbance, movement, spatial orientation, dead-reckoning

Reservoir rodent host population dynamics drive wildlife-originated zoonotic infections in humans

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Wildlife-originated zoonoses, diseases that are transmitted from animals to humans, pose a major risk to human health. Rodents are important reservoir hosts of many zoonotic pathogens, and rodent population dynamics affect the infection dynamics of rodent-borne diseases, such as diseases caused by hantaviruses. However, the role of rodent population dynamics in determining the infection dynamics of rodent-associated tick-borne diseases, such as Lyme borreliosis (LB), have gained limited attention. Here, we quantify the associations between multiannual rodent density fluctuations and laboratory-diagnosed LB and Puumala Orthohantavirus (PUUV) infections by using 25-year (1995-2019) monthly bank vole (*Myodes glareolus*) and human infection data in Finland. In addition, we examined nine years data on LB cases, diagnosed based on symptoms. Both bank vole abundance as well as LB and PUUV infection incidence in humans showed approximately 3-year cycles. Directly transmitted PUUV infections followed the bank vole host abundance fluctuations with two-month time lag, whereas tick-transmitted LB was associated with bank vole abundance *ca.* 12 and 24 months earlier. However, the strength of association between bank vole abundance and LB incidence with 12-month time lag varied over the study years. Our results indicate that not only human risk to acquire PUUV, but also LB is driven by multiannual reservoir rodent host population fluctuations.

Influence of handler relationships and experience on the health, stress and behaviour of semi-captive Asian elephants

Jennifer Crawley

Declining wild populations combined with accumulating captive populations of e.g. livestock, pets, draught and zoo animals, have resulted in some threatened species with a substantial proportion of their populations in captivity. The interactions animals have with humans within captivity considerably impact their wellbeing, and effects depend on the familiarity and quality of the relationship. Asian elephants have been managed by humans for millennia, and >25% live in captivity today, mostly in their range countries with one or more handlers (mahouts) almost entirely responsible for their care. Few studies have investigated these relationships, or the impact of shifts seen in handling systems across Asia. Here, I will talk about a study on a population of semi-captive Asian elephants with detailed handling records and veterinary management, allowing assessment of multiple welfare indicators in relation to mahout-elephant relationship lengths and mahout total experience. We measured elephants' physiological stress (Faecal glucocorticoids, Heterophil:Lymphocyte ratio), muscle damage (Creatine Kinase/CK), immunological health (Total White Blood Cell count/TWBC), and behaviour (response to simple commands). An elephant's CK, and to some extent TWBC, increased logarithmically with their mahout's total experience, indicating muscle damage and inflammation. The effect of relationship length on CK was age dependent, declining with longer mahout-elephant relationships after the age of 18. Elephants also showed behavioural distinctions, responding more to familiar mahouts and faster to mahouts they had known for longer. Our results suggest little evidence of the mahout-elephant relationship impacting physiological stress but mahout experience is linked to physiological responses, and elephants require behavioural adjustment periods following mahout changes.

How Human-Animal relationship affects the behavioural response in a working context: The Asian elephant as a study case.

Oceanne Liehrmann

The nature of relationships between people and animals has been of interest for many centuries. However, the experimental study of these interactions is a relatively recent development, despite the suggestion that Human-Animal interactions may have consequences for animal welfare. Intriguingly, although the Human-Animal relationship is an important factor to take into account in animal management, researchers have to date overlooked some animals: the working animals for hard labour known as draught animals. To investigate the human-Draught Animal relationship,87 Myanmar timber elephants were asked to respond to the call of their own mahout (elephant caretaker) or the call of another mahout, and 52 of them were asked to walk on a novel surface. These tests aimed to assess if a long-lasting relationship between handlers and animals affects the quality of the response to work orders in a usual context or a disturbed context (confronted with novelty). The analysis revealed that most of the elephants responded only to their own mahout and elephants responding were less disturbed by the novel surface when they had a long-lasting relationship with the mahout calling. The success rate was also driven by the age of the elephant in interaction with the mahout's identity, indicating the importance of training and for the animal and handlers to know and understand each other. On the contrary, when novelty was involved, the older elephant trend to failed more following the common theory of young individuals being less neophobic than older ones. This study is the first to experimentally assess the relationship between handlers and draught animals, highlighting its importance in improving work quality and performances. Further research is needed to investigate the role of the Human-Draught Animal relationship in terms of animal welfare and handlers' security.