

# The 11<sup>th</sup> Polish Evolutionary Conference

8-10 September 2025 Warsaw, Poland

**PROGRAM & ABSTRACT BOOK** 

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Faculty of Biology, University of Warsaw The Committee of Environmental & Evolutionary Biology, Polish Academy of Sciences







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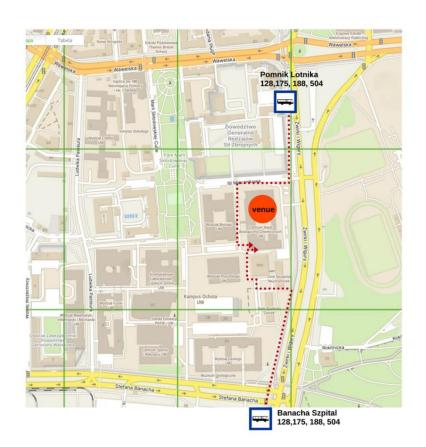
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# **VENUE**

Campus "Ochota", Centrum Nauk Biologiczno-Chemicznych Uniwersytetu Warszawskiego (CNBCh) ul. Żwirki i Wigury 101, 02-089 Warszawa.

Campus Ochota is served by bus lines 175 (Airport to the city centre via Central Railway Station), 128 and 504 from the city centre (including Central Railway Station) 188 from the airport. Nearest bus stops are called "Pomik Lotnika" (when travelling from the city centre) or "Banacha-Szpital" (when travelling from the airport).

# **PROGRAMME**

| MONDAY 8.09             |                           |  |
|-------------------------|---------------------------|--|
| from 09:00              | Registration              |  |
| 11:45-12:00             | Opening                   |  |
| Session: Archeo & Paleo |                           |  |
| 12:00-13:00             | plenary: Michael Westbury |  |
| 13:00-14:00             | lunch                     |  |
| 14:00-14:20             | Alicja Kaźmierkiewicz     |  |
| 14:20-14:40             | Anita Murawska            |  |
| 14:40-15:00             | Jonatan Audycki           |  |
| 15:00-15:30             | coffee break              |  |
| Session: Anthropocene   |                           |  |
| 15:30-15:50             | Sabina Nowak              |  |
| 15:50-16:10             | Robert Mysłajek           |  |
| 16:10-16:30             | Amelia Chyb               |  |
| 16:30-16:50             | Guillaume Wos             |  |
| 16:50-17:20             | coffee break              |  |
| 17:20-17:40             | Barbara Płaskonka         |  |
| 17:40-18:00             | Ignacy Stadnicki          |  |
| 18:00-18:20             | Marcin Mazurkiewicz       |  |
| 18:20-18:40             | Yuan Fu Chan              |  |

| TUESDAY 9.09                      |                                       |  |
|-----------------------------------|---------------------------------------|--|
| Session: Genomics and adaptations |                                       |  |
| 9:00-10:00                        | plenary: Lewis Stevens                |  |
| 10:00-10:30                       | coffee break                          |  |
| 10:30-10:50                       | Viktor Kovalov                        |  |
| 10:50-11:10                       | Abosede Olarewaju                     |  |
| 11:10-11:30                       | Barsa Das                             |  |
| 11:30-11:50                       | Małgorzata Pilot                      |  |
| 11:50-12:20                       | coffee break                          |  |
| 12:20-12:40                       | Yellapu Srinivas                      |  |
| 12:40-13:00                       | Wiesław Bogdanowicz                   |  |
| 13:00-13:20                       | Clément Car                           |  |
| 13:20-13:40                       | Joanna Kołodziejczyk                  |  |
| 13:40-14:40                       | lunch                                 |  |
| 14:40-15:40                       | plenary: Krystyna Nadachowska-Brzyska |  |
| 15:40-16:00                       | Nermeen R. Amer                       |  |
| 16:00-16:20                       | Bartłomiej Molasy                     |  |
| 16:30-18:00                       | Welcome Reception / Poster session    |  |

|   | WENDESDAY 10.09                     |  |
|---|-------------------------------------|--|
| WENDESDAY 10.09                                   |                                     |  |
| Session: Symbiosis                                |                                     |  |
| 9:00-10:00  | plenary: Gytis Dudas                |  |
| 10:00-10:30                                       | coffee break                        |  |
| 10:30-10:50                                       | Metody Hollender                    |  |
| 10:50-11:10                                       | Daniel Méndez-Sánchez               |  |
| 11:10-11:30                                       | Veronika Andriienko                 |  |
| 11:30-11:50                                       | Ivan Garcia Cunchillos              |  |
| 11:50-12:20                                       | coffee break                        |  |
| Session: Sexual selection and life history traits |                                     |  |
| 12:20-12:40                                       | Julia Kandulska                     |  |
| 12:40-13:00                                       | Neelam Porwal                       |  |
| 13:00-13:20                                       | Mohammadjavad Haghighatnia          |  |
| 13:20-13:40                                       | Ludwik Gąsiorowski                  |  |
| 13:40-14:00                                       | Arkadiusz Fröhlich                  |  |
| 14:00-15:00                                       | lunch                               |  |
| Session: Ecophysiology                            |                                     |  |
| 15:00-15:20                                       | Jakub P. Płachta                    |  |
| 15:20-15:40                                       | Julita Sadowska                     |  |
| 15:40-16:00                                       | Andrzej K. Gębczyński               |  |
| 16:00-16:30                                       | coffee break                        |  |
| 16:30-16:50                                       | Małgorzata M. Lipowska              |  |
| 16:50-17:10                                       | Martyna Jankowska-Jarek             |  |
| 17:10-17:40                                       | Tomiałojć medal talk                |  |
| 17:40-18:00                                       | Best talk/poster prizes and closing |  |

# PLENARY SPEAKERS

#### Dr hab. Krystyna Nadachowska-Brzyska

Jagiellonian University, Krakow, Poland

Krystyna Nadachowska-Brzyska has been working on diverse evolutionary biology topics including speciation with gene-flow in amphibians and birds, variation of major histocompatibility genes in several newt species, historical effective population size changes and many more. Currently, she investigates the influence of extreme fluctuations in population size of outbreaking species on the dynamics of neutral and adaptive genetic variation using genome-wide datasets and novel analytical approaches.



# Dr. Gytis Dudas

Life Sciences Center, Vilnius University, Lithuania

Gytis Dudas studies RNA virus evolution at both short (genomic epidemiology) and long (metagenomic virus discovery) timescales.



# Dr. Lewis Stevens

Wellcome Sanger Institute, UK

Lewis Stevens is an evolutionary biologist within Prof. Mark Blaxter group. He is interested in genome evolution in animals, with a particular interest in nematodes.



### Prof. Michael Westbury

University of Copenhagen, Denmark

Michael Westbury's research focus lies in the production and analysis of genomic data from modern, historical, and palaeontological specimens. Throughout his career he has been studying a variety of (mainly) large mammalian species by utilising population and comparative genomic analyses to better understand their respective evolutionary histories and relationships.



#### Krystyna Nadachowska-Brzyska

Going beyond SNPs in adaptation genomics - the role of inversions in Europe's most destructive forest pest

The diversity of life can manifest itself at many different levels, including the diversity of intraspecific variation in genomic architectures. Among these, polymorphic inversions are often found to underlie complex phenotypic polymorphisms and facilitate local adaptation in the face of gene flow. In addition, a growing body of research is providing evidence for genomes with dozens of large inversions segregating within species, raising questions about the extent of inversion landscape complexity, the evolutionary forces that maintain/influence such diversity, and our ability to account for this complexity in evolutionary inference. I will begin my presentation with a brief introduction to adaptation genomics and structural variation, followed by an overview of our research on the spruce bark beetle. We recently discovered that the spruce bark beetle genome harbors 30 large polymorphic inversions spanning ~30% of the genome. These inversions vary in size and in the degree of recombination suppression, are highly polymorphic across the species' range, and frequently overlap, creating a complex genomic architecture. Our research focuses on two main questions:

- Why are so many inversions maintained within a species' genome, and what evolutionary forces contribute to their persistence?
- Do inversion-rich genomes bias evolutionary inference, such as tests for selection or demographic analyses? Our results suggest that inversions are maintained by multiple, often non–mutually exclusive evolutionary forces that are difficult to disentangle, and that their presence can influence evolutionary inference in diverse ways.

#### **Gytis Dudas**

More questions than answers: the Wuhan mosquito virus 6 pandemic in mosquitoes

Wuhan mosquito virus 6 (WuMV-6) is a globally distributed orthomyxovirus (RNA virus family that includes influenza viruses) of Culex mosquitoes exhibiting rapid migration and high rates of non-synonymous evolution in its surface protein gp64. These features would typically indicate a virus capable of infecting a vertebrate host that would both disperse it across large distances and exert a selective pressure for amino acid replacement via neutralising antibodies. However, despite years of extensive bioinformatic and experimental analyses, this hypothesis seems less and less likely. In this talk, I'll walk through the knowns and unknowns of WuMV-6 as a study system and discuss the unfamiliar ecology and evolution of arthropod RNA viruses.

#### **Lewis Stevens**

What sequencing the genomes of individual worms can tell us about parasite biology and evolution

Host-parasite interactions are powerful drivers of evolutionary change, often leading to extreme genetic diversity in genomic regions directly involved. Parasitic nematodes are ubiquitous in natural ecosystems, cause immense harm to livestock and crops, and infect more than 1.5 billion people worldwide. A central guestion is how these organisms survive in the face of host immune defences. Recent advances in genome sequencing, particularly longread technologies, now permit characterisation of the full spectrum of genetic variation within a species, from single nucleotide variants to large structural variants, known as the 'pangenome'. However, the application of these technologies to parasitic nematodes has been hindered by the fact that individual parasites contain very limited amounts of DNA. In this talk, I will show how new low-input protocols are allow us to generate high-quality genomes from individual parasites. Using these approaches, we have discovered that parasitic nematode genomes contain large regions that differ markedly between individuals of the same species, termed hyperdivergent haplotypes (HDHs). Strikingly, many HDHs encode genes implicated in host interaction, including antigens trialled as vaccine candidates, and frequently comprise distinct sets of functionally related genes. I hypothesise that this extreme genetic diversity plays a central role in parasite survival by allowing these parasites to evade the host immune response. I will discuss the implications of these findings, particularly for anti-parasite vaccine development. I will also discuss how single-parasite sequencing can be scaled up to provide new insights into parasite evolution, ecology, and their capacity to cause harm.

### Michael Westbury

Palaeogenomics as a tool to understand evolution, extinction, and the future of biodiversity

The recovery of DNA from ancient remains has revolutionised evolutionary biology. By combining ancient DNA with modern genomic approaches, palaeogenomics enables us to reconstruct evolutionary histories, trace genetic change through time, and investigate extinct species in unprecedented detail. In this talk, I will highlight how palaeogenomics has provided novel insights into the rise and fall of extinct species such as the blue antelope, cave hyena, and the giant deer. I will also show how palaeogenomic data from extant species, in this case bowhead whales, provides a window into their future by showing how populations responded to past climatic shifts, habitat changes, and human pressures, and how we can use that going forward in time. Together, these studies demonstrate how understanding past dynamics of adaptation, connectivity, and decline not only illuminates the evolutionary past but also provides critical perspectives for predicting the resilience and vulnerability of species facing current and future environmental change.

# ABSTRACT BOOK

--- TALKS ---

ABSTRACTS: TALKS SESSION: ARCHEO & PALEO

Ancient and modern genomes of the common vole, *Microtus arvalis* (Rodentia, Cricetidae), reveal the complex evolutionary history of Western Carpathian populations

### Alicja Kaźmierkiewicz<sup>1</sup>

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Microtus arvalis is a rodent species widespread in Europe present and past. In much of Central Europe extant population belongs to the Eastern mtDNA lineage. Previous studies have identified a Central mtDNA lineage in the same region in the Late Pleistocene and Early Holocene suggesting a population turnover at the Pleistocene/Holocene transition. However, the course of this replacement remained unclear. To further understand the evolutionary history of the common vole we generated 13 late Pleistocene and early Holocene low coverage genomes from paleontological sides across Western Carpathians, and 39 mid- to high- coverage modern genomes from Central Europe. Principal Component Analysis (PCA) of newly generated and previously published data revealed two main clusters: one comprising modern samples belonging to Eastern mtDNA lineage, and the other containing ancient samples from both Eastern and Central mtDNA, and a few modern Eastern samples. This suggests that the expanding Eastern lineage mixed with the local Central population; however, traces of this admixture are not readily visible in the extant population. These results raise several questions, including the reasons for population expansion into areas occupied by other population of the same species, the potential advantageous adaptations of the Eastern population to the environment, whether the signal of such adaptations can be detected in the genome, and why do modern populations in the Western Carpathians show no trace of the Central population. Overall, preliminary results of our project suggest a more complex evolutionary history of the common vole in the Western Carpathians than previously speculated.

# Multi-Proxy Reconstruction of Long-Term Vegetation and Fire History at the Lipsk Archaeological Site, NE Poland

**Anita Murawska**<sup>1</sup>, Monika Chmielewska<sup>1</sup>, Michał Sulwiński<sup>1</sup>, Marcin Frączek<sup>2</sup>, Michał Przeździecki<sup>3</sup>, Beliavets Vadzim<sup>4</sup>, Adam Wawrusiewicz<sup>5</sup>, Krzysztof Żurek<sup>5</sup>, Monika Mętrak<sup>1</sup>, Małgorzata Suska-Malawska<sup>1</sup>.

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Human activity has long shaped natural environments, with especially transformative impacts during the Mesolithic-Neolithic transition. This period saw a major shift from mobile hunter-gatherers to more sedentary agricultural societies, leading to lasting landscape changes. However, human-environment interactions during the Sub-Neolithic period remain less understood, particularly regarding semi-nomadic cultures who adopted pottery but maintained foraging lifestyles. This study investigates changes in vegetation type and the occurrence of fire episodes, which might be linked to the Sub-Neolithic Neman culture at a unique site in Lipsk, northeastern Poland. The Lipsk site is a dune surrounded by extensive peat deposits, creating an exceptional natural archive preserving detailed records of past environments. The Neman culture, present during the Sub-Neolithic, were semi-nomadic hunter-gatherers, integrating pottery use. Their activities, including possible landscape management through fire, may have left detectable traces in the peat stratigraphy. Understanding how these communities influenced their environment contributes to broader discussions on the origins of possible prehistoric human-driven landscape changes.

To explore this, we applied a multidisciplinary palaeoecological approach combining biomolecular and microscopic analyses. Gas chromatography-mass spectrometry (GC-MS) was used to analyse n-alkanes extracted from peat layers. These molecules serve as biomarkers to reconstruct past vegetation and identify changes potentially linked to human activity. Additionally, microscopic analysis of charcoal particles in the peat provides direct evidence of local fire events, which may reflect human-induced burning.

Our results will offer valuable insights for both palaeoecology and archaeology, contributing to a better understanding of the possible prehistoric human-nature relations. The multi-proxy analyses are scheduled to be completed by June 2025, followed by a comprehensive data interpretation. This study highlights the importance of performing multi-proxy interdisciplinary studies, combining biology and archaeology to detect subtle early anthropogenic impacts that may not appear in traditional archaeological records.

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<sup>&</sup>lt;sup>5</sup>The Podlaskie Museum, Białystok, Poland

ABSTRACTS: TALKS SESSION: ARCHEO & PALEO

# A new austrolimulid from the Triassic of Poland presents insight into xiphosurid evolution and palaeobiogeography at the dawn of the Mesozoic

**Jonatan Audycki**<sup>1</sup>, Russell D. C. Bicknell<sup>2</sup>, Grzegorz Niedźwiedzki<sup>3,4</sup>, Kenneth De Baets<sup>1</sup> <u>j.audycki@uw.edu.pl</u>

Xiphosurans, commonly known as horseshoe crabs, are predominantly marine chelicerates famous for their apparent morphological stasis and limited diversity. However, some fossil representatives of this group have radically diverged in form from the classic horseshoe crab morphology emphasized by the Recent Limulus, challenging this traditional view of horseshoe crab evolution and giving rise to questions about the possible drivers of such changes. This is especially true for the austrolimulids—a group of horseshoe crabs known almost exclusively from the Triassic, which developed extremely elongated genal spines and have been sometimes linked to brackish and freshwater environments. Here, we report new horseshoe crab specimens from the lower part of the Middle Buntsandstein deposits in Holy Cross Mountains (southern Poland) that most likely constitute a new genus within the Austrolimulidae. Dated to the latest Induan/earliest Olenekian, the new taxon seems to represent a transition between a less-derived anatomy of geologically older austrolimulids and the extreme morphology of Austrolimulus fletcheri from the Middle Triassic of Australia, which is supported by a new geometric morphometry analysis. This find provides an important insight into the distribution of morphologically 'extreme' austrolimulids and supports their possible connection with brackish habitats, making it a favorable starting point for further studies on their evolution and ecology. Additionally, based on palynological insight and analysis of associated fossils, we provide further support for the latest Induan-earliest Olenekian marine influence in the Polish Basin.

#### **FUNDING**:

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# Wolves as indicators of farmers' adherence to health regulations in Poland

#### Sabina Nowak<sup>1</sup>

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Carcasses of farm animals, if not properly utilized, may be a source of viral, bacterial, and parasitological diseases harming humans, as well as wild and domestic animals. Furthermore, carcass deposition near human settlements may attract wildlife and escalate conflicts. Therefore, monitoring farmers' compliance with sanitary laws is instrumental in sustaining long-term human-wildlife coexistence. We illustrate the sentinel potential of wolves, analyzing the content of their scats and using GPS tracking to assess farmers' compliance with EU sanitary regulations regarding the utilization of livestock and poultry carcasses. We performed a systematic review of all papers focused on the wolf diet based on scat analysis in forests and additionally collected and analyzed the content of wolf scats from human-dominated landscapes (23 study areas, >5700 scats) to reveal the share of domestic animals in the wolf diet across the habitat gradient in Poland. Furthermore, we followed 23 wolves equipped with GPS-GSM collars to indicate the species, locations, and sources of domestic animals they consumed. We also installed camera traps near two poultry farms to obtain detailed data on the wolves' presence in dumps where carcasses were deposited. We revealed that in Poland, domestic animals comprised, on average, 3.3% of consumed biomass (range 0-14.2%). The amount of domestic animals in the wolf's diet is not, however, correlated with the area of farmlands within study areas. We revealed that collared wolves can find and forage on carcasses of livestock that have been illegally dumped in forests and marshlands. Wolves were able to dig up farm animal carcasses buried in earthen pits. We also observed that wolves, especially young individuals, may regularly visit the vicinity of poultry farms, forage on the remains of illegally dumped birds, and rest on manure piles, which causes severe dermatosis and lowers their fitness. Our study shows the potential of GPS-tagged wolves as sentinels for monitoring farmers' compliance with sanitary laws.

# Patterns of the wolf mortality on roads in Poland

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The recovery of the wolf population across human-dominated landscapes of central Europe coincides with an increasing mortality of wolves on roads, the level of which remains unknown. In Poland, where the species has been strictly protected since 1998, and its current range covers forests and mosaics of natural and humanmodified areas, we opportunistically collected data on wolves hit by vehicles from 2002 to 2023, revealing their distribution, seasonal variation, sex and age structure, social status and road category at the kill site. We also conducted GIS analyses of habitats along the road and their impact on the risk of collision. Datasets from 20 wolves collared in 2014-2023 were used to assess the number of crosses per wolf per day, time of crosses, and traffic on the road section, differences in road crossings between wolf age categories, and risk of being killed on the road. Within 20 years, we recorded 447 wolves struck by vehicles on roads. Most of them died between 2018 and 2023. Among road casualties with defined age classes, there were more adults (> 1 year, 69%) than juveniles (< 1 year). Adults were killed mainly in late autumn (41% of adults' mortality) and then in late winter (23%). Juveniles died on roads mostly in autumn, with 61% of pups' mortality. Mainly young (1-3 years) adult wolves were killed on roads (70%), much fewer (20%) wolves over 3 years old, and only 10% older than five years. The youngest road-killed pup was seven weeks old, but the most frequent were those of six and seven months old, making 36 % of all pups' deaths. Wolves mainly died on national (47%) and regional roads (29%). Most roadkills (78%) were found on road sections crossing forested areas, at least on one roadside (78%), where traffic was below 9.000 vehicles/per day. Wolves with GPS-GSM collars crossed roads on average 2.6 times per day (range 0.7 – 7.8), mainly during the night (88%) between 8 pm and 8 am. Our study results can help create solutions to reduce wildlife collisions and assess the impact of traffic on wolf populations.

## Don't be shy – how urban life modulates behaviour in a common waterbird

**Amelia Chyb**<sup>1</sup>, Radosław Włodarczyk<sup>1</sup>, Joanna Drzewińska-Chańko<sup>1</sup>, Damian Andrysiak<sup>1</sup>, Jan Jedlikowski<sup>2</sup>, Piotr Minias<sup>1</sup>

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Urbanization processes can markedly affect different aspects of animal behaviour via alterations in environmental conditions, intensive human disturbance and relaxation of predator pressure. Individuals from urban bird populations often present specific behavioural phenotypes, such as elevated aggression, increased boldness, decreased neophobia, and reduced fear of humans (so called 'urban wildlife syndrome'). Here, we investigated behavioural responses to the appearance of a human intruder (shyness) and a novel object (neophobia) at the nest in three populations of the Eurasian coot *Fulica atra*. Specifically, we compared behaviour of coots between different levels of urbanization (urban populations vs nonurban population) and between populations showing contrasting history of urban colonization (recently-established vs long-established urban population). We found that individuals from long-established urban population required shorter time to return to the nest and resume incubation after the appearance of a human intruder (i.e. showed reduced shyness) than nonurban ones. Similarly, they also had higher probability of returning to the nest and resuming incubation under exposure to a novel object at the nest (i.e. showed reduced neophobia). Coots from recently-established population showed intermediate responses in both experiments. Our results show that the effects of urbanization on avian behaviour may be complex and dependent not only on specific environmental conditions prevailing in human-dominated areas, but also different histories of urban colonization.

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# Human activities drive phenotypic variations at a microgeographic scale in the damselfly *Ischnura elegans*

#### Guillaume Wos1

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Urbanization has emerged as one of the main drivers of habitat transformation affecting ecosystem functioning. Urbanization has a profound impact on organisms and may be the source of important phenotypic changes. Moreover, the patterns of phenotypic variation do not depend only on the level of urbanization but also on other environmental features e.g., percentage of forested habitats or crops, affecting, among other things, connectivity between habitats and dispersal ability of organisms.

In the present study, we investigated the phenotypic variation related to urbanization measured as the percentage of impervious surface in approx. 800 adult blue-tailed damselflies *Ischnura elegans* sampled across 22 water bodies in Poland spanning different urban and rural environments also characterized by various levels of agricultural land, forested area and water parameters related to human activities (pH and salt concentration). Adults were screened for traits related to size (head with, thorax length, femur length) and dispersion (wing size and shape). For a subset of the ponds sampled, we also investigated the effects of the environment on size, growth and development time on their offspring (larval stage) raised in growth chambers under controlled conditions.

We found that urbanization significantly affected adult size with urban individuals being in general smaller, but the effects of urbanization were also dependent on other parameters e.g., salinity and the percentage of forested habitat. Wing size was more affected by urbanization than wing shape, with bigger wings in rural environments combined with low levels of salinity. The effects of urbanization were also sex-specific with urban males having the smallest size.

For their offspring, urbanization and the percentage of forested habitat significantly impacted larval survival with higher survival in urban and open habitats. The shortest larval development time was found in rural and forested environments.

In conclusion, our results provide evidence that urbanization interacted with other environmental parameters to shape adult phenotype that may ultimately affect their dispersal ability and fitness. Furthermore, the effects of urbanization were also found in their progeny suggesting a potential genetic basis of the phenotypic variation and/or maternal effects.

# Pollination on the Fly: urban pollen transfer on late flowering ivy

#### Barbara Płaskonka<sup>1</sup>

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Cities are complex and often very demanding habitats for wild organisms, including plants and pollinating insects. Available research indicates the important role of urban areas in biodiversity protection, however, often overlooks the important role of the urban flies in its maintenance. We tracked quantum dots-labeled pollen grains transfer between four patches of Hedera helix, a late-season flowering species, where flies are one of the most frequent flower visitors. The mosaic of green and impermeable areas in cities may influence the movement of pollinators between attractive populations of flowering plants and our study was the first attempt to track pollen grain movements in a pollinator assemblage dominated by flies. Our study shows that the frequency of pollinator visits and pollen transfer were related to the proportion of green areas and umbel density, while pollen transfer was also influenced by distance between populations. Results show that pollen is transferred between small isolated ivy populations due to pollinator movement, indicating that these populations may serve as stepping stones for pollinators among patches of urban greenery. This is especially important due to the lack of food sources in the late growing season.

# An analysis of passerine egg traits variation across the urban mosaic: Birds in the city invest in the quality of eggs over quantity

### Ignacy Stadnicki<sup>1</sup>

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- 1. Rapid urbanisation is a recent worldwide phenomenon. Compared to natural areas, cities are characterised by a number of unique factors affecting both abiotic and biotic elements of ecosystems, ultimately changing the ecoevolutionary dynamics of those habitats. While urban birds differ in morphology, behaviour and reproductive patterns, the impact of urbanisation on egg traits a key aspect of the avian life-cycle remains underresearched. Given the limited availability of small snails (a primary source of calcium for laying females) in cities, eggs from heavily urbanised areas were predicted to be smaller and more pigmented, indicating thinner shells and limited resources.
- 2. In order to better understand how urbanisation might affect eggs, data on 751 great tit eggs from 91 clutches, spread across 8 study sites in a city mosaic, were collected for two breeding seasons. All clutches were photographed using a standardised approach, and further quantified using visual scoring and digital imaging methods to assess egg volume and pigmentation patterns. Urbanisation was quantified in the vicinity of each clutch via satellite imagery. Statistical analyses were performed using repeatability tests, ANOVA and GLMMs with egg traits as response variables, and environmental and life history factors as explanatory variables.
- 3. Contrary to the original predictions, egg traits correlated with life history traits, but not with urbanisation. Covariation with life history traits here aligned with earlier findings of egg size and pigmentation research conducted on natural populations. While, the lack of correlation with urbanisation aligns with limited research on urban populations. Moreover urban clutches had fewer eggs, a trend that was reported in a number of previous studies.
- 4. In this study, great tit egg traits did not differ between natural and urban populations, yet urban clutches were consistently smaller. In the light of these results, we suggest that birds in the city invest in the quality of eggs over the quantity. Such trade-off may allow for successful reproduction at this stage of breeding, enabling birds to cope with urban-driven environmental constraints (likely by increasing their calcium foraging efforts), but only to a certain extent.

## The impact of forest management on functional diversity of bryophytes in close-to-natural forest

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The impact of forest management practices, although well studied in the context of increasing the efficiency of forest production and their influence on endangered and threatened species, still leaves substantial knowledge gaps regarding forest ecosystem functioning. Close-to-natural forests, as remnants of once vast forest complexes, are particularly valuable due to their long-term exemption from intensive human activity. They are characterized by a distinct composition of plant groups, sometimes closely tied to this type of forest. Among them, bryophytes — a key group — contribute significantly to both the taxonomic and functional biodiversity of forests. Their strong association with diverse microhabitats and high sensitivity to environmental factors make them one of the first organisms to respond to environmental changes. Thus, they serve as effective indicators of ecological shifts caused by climate change and anthropogenic disturbance. In this study, we investigated the impact of forest management on the functional diversity of bryophytes in oak-hornbeam (Tilio-Carpinetum) forests. Research was conducted in the Białowieża Forest (NE Poland), in two contrasting areas: the strictly protected Białowieża National Park and an adjacent managed forest. We surveyed 120 square-shaped plots (51 in the Park and 69 in the managed forest), where oak-hornbeam forest was predominant. Bryophyte species were recorded across all available substrates—living trees, deadwood, rocks, soil, and others. We also assessed species frequency in each plot. We recorded 126 bryophyte species: 113 in the National Park and 93 in the managed forest. Using ordination analysis, we found that forest management strongly affects bryophyte communities, reducing their biodiversity. We further examined functional traits, including size, growth forms, life forms and strategies, and functional diversity metrics. Our results indicate that bryophyte communities in forests minimally impacted by humans show greater functional diversity and functional divergence. Furthermore, communities in managed forests exhibit traits of environmentally homogenized habitats while experiencing increased ecological stress due to altered habitat conditions. This study offers new insights into the effect of forest management on forest ecosystem functioning. By focusing on bryophytes — an ecologically sensitive yet understudied group — we contribute to a broader understanding of the functional and phylogenetic scope of human impact on forest biodiversity.

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## How urbanisation shapes the evolutionary response of the wild radish Raphanus raphanistrum

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Urbanisation leads to the transformation of abiotic and biotic environments in ways that might mediate the evolutionary adaptation of organisms. However, the impact of urbanisation on evolutionary processes in populations remains poorly understood. Here, we investigate how urbanisation shapes the evolutionary response of the wild radish Raphanus raphanistrum. We conducted field surveys across the southern Italian Campania region and a common garden experiment to examine the phenotypic divergence between urban and natural populations. A genome-wide association study (GWAS) and population genetic analysis were also performed to investigate the genetic basis of urban adaptation. Our results show that urban populations had significantly smaller flowers and earlier flowering times compared to natural populations. Selection analyses indicate that natural selection generally favours earlier flowering times and longer petal lengths. However, a trade-off between these traits suggests habitat-specific selection: earlier flowering times were favoured in urban environments, while larger flowers were selected in natural ones. Population genetics analysis reveals low genetic differentiation and frequent gene flow among urban and natural populations. The GWAS identifies 32 single-nucleotide polymorphisms (SNP) significantly associated with urbanisation, 21 of which are located within gene regions. Notably, one of these genes, AT3g27390 is linked to the transition to reproductive development. Overall, our findings highlight how natural selection in urban contexts shapes rapid adaptive phenotypic divergence in R. raphanistrum despite the presence of gene flow.

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# Triploidy in parasitic worms: an overlooked aspect in host-parasite dynamics and evolution of drug resistance?

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Parasitic worms affect human health, infect livestock, and cause crop losses, making them important research target. Most studies assume that parasitic worms are diploid and reproduce sexually, as sexual selection is important to counter evolution of resistance in hosts. However, some species, like *Paragonimus westermani*, have triploid individuals that reproduce parthenogenetically. This raises an important question about the role of triploidy and asexuality in parasite evolution. Yet, to our knowledge, no study has investigated the prevalence of triploidy and its associated reproductive modes. We address this gap by conducting a systematic review on triploidy and reproduction in parasitic worms.

We searched for literature on Web of Science and Scopus databases using keywords related to triploidy and taxonomy of parasitic worms from Platyhelminthes and Nematoda. In addition, we included keywords covering free-living flatworms for comparison. After removing duplicates, we screened 416 papers and retained 100 relevant studies. From these, we extracted species names, reproductive modes, coexistence with other ploidy levels, and origin of triploidy.

We found 54 triploid species, out of which 30 are parasitic: 38 flatworms (16 parasitic) and 16 roundworms (14 parasitic). Among parasitic triploids are such species as *Fasciola hepatica* and *Schistosoma* mansoni, species important for human and livestock health, as well as root-knot nematodes from *Meloidogyne* genus. Two triploid species (*Haemonchus contortus* and *S. mansoni*) are known to have drug-resistant populations, although links to triploidy remain unclear. Most triploids reproduce asexually, though some free-living flatworms can switch to fully functional sexual reproduction.

This study is the first systematic approach to review triploidy in parasitic worms. Low number of species with triploidy compared to the total number of species may reflect either rarity of triploidy or potential research bias. Thus, with our study, we want to draw attention to the phenomenon of triploidy and encourage further research testing how combination of increased genome size and asexuality affect evolution of parasitic worms.

# Phylogeography reveals the evolutionary histories of a co-evolving host-parasite system

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Post-glacial recolonisation of Europe by hosts and their parasites has led to spatial genetic differentiation caused by migration and local adaptation. Incorporating parasites' phylogeographic information in the study of hosts can explain unknown spatial differences in the genetic diversity of their hosts. However, the impact of both migration and adaptation on genome-wide variation is still poorly understood especially in parasites. In this study investigates the spatial genetic variation in the bank vole (*Myodes glareolus*) and its most common nematode parasite, *Heligmosomum mixtum*. Using quaddRAD sequencing, we genotyped 318 *M. glareolus* and 236 *H. mixtum* from 17 and 11 populations respectively from Poland, Czech and Finland. We obtained 35,522 and 6,464 variant sites in the host and in the parasites. Our findings reveal that the parasite population is more structured compared to its host, with Czech populations being more highly genetically differentiated and gene flow in the host does not correspond to gene flow in the parasites. These results show how host movement, parasite dispersal, and local adaptation interact to shape genetic patterns, and highlight the importance of including parasite genomics in coevolutionary studies.

# Investigating a possible hybridisation/introgression case in flightless Central Asian longhorned beetle (Coleoptera: Cerambycidae)

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The flightless longhorned beetle genus *Eodorcadion* belongs to the highly diverse and taxonomically complex tribe Dorcadionini s.l. *Eodorcadion* is largely distributed in the vast areas of Central Asia, including the whole territory of Mongolia. Previous studies on the group assumed that one of the reasons behind such huge taxon richness in Dorcadionini s.l. is hybridization and sometimes subsequent introgression.

We diagnose a potential case of hybridisation followed by introgression between *E. exaratum*, *E. kozlovi*, and *E. oryx*, the latter being presumably the introgressed taxon of the former two species. In this study, we focus on *E. oryx* and its molecular affinity for the putative parental species.

Although described as separate species, the discordance between morphological characters and molecular data suggests a more complex evolutionary history between the concerned species. While *E. oryx* is morphologically distinguishable, in mtDNA (COI, cytb), the examined individuals were identical to one another, being also very similar to *E. kozlovi*, while in nDNA (CAD, ITS2, Wng), individuals of *E. oryx* were surprisingly different from each other, much more than in the case of two relatively closely related species, and some of them were identical to *E. exaratum*.

We hypothesize that *E.oryx* was formed as a result of hybridisation between *E. exaratum* and *E. kozlovi*, and subsequent introgression with *E. kozlovi*. The known geographic range of *E. oryx* which is separated from both the related species is possibly a key reason for its stability as an individual taxon. To lay more evidence to this claim, we compared both mtDNA and nDNA sequences of E. oryx with both the related species across their distribution range. Revealing the evolutionary relationships between the three species and analysing the obtained sequences individually will lead us to decipher factors affecting species formation through hybridisation and introgression and their stability in time and space.

By resolving hybridization-driven discordance in *Eodorcadion* beetles, we illuminate how introgression and geographical isolation shape species boundaries. This work advances our understanding of speciation mechanisms in rapidly radiating lineages with complex evolutionary histories.

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## Environmental drivers of domestic dog introgression in wild canids

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Introgressive hybridisation between wild canids and free-ranging domestic dogs is a widespread phenomenon with important ecological and evolutionary consequences. Multiple studies hypothesised that hybridisation rate is accelerated by anthropogenic habitat modifications, but this hypothesis has never been tested. We investigated environmental factors that contribute to the presence and proportions of dog ancestry in populations of three canids with varying evolutionary distance from dogs: dingoes, grey wolves and golden jackals, across their Australian and Eurasian ranges, respectively. We quantified dog ancestry at the chromosomal level in individual canids based on genome-wide single nucleotide polymorphisms. For each genotyped individual, we collected environmental data related to climate, topography, vegetation, land cover and anthropogenic landscape modifications to identify key environmental drivers of dog introgression using Random Forest models. We found that climatic variables and the Human Footprint Index were significant predictors of introgression in all three wild canid taxa. Climatic predictors were taxon-specific and their impact may be associated with their influence on the relative densities of wild canids and free-ranging dogs. In contrast, the positive association between the Human Footprint Index and introgression rates was consistent across all three canids, implying that anthropogenic landscape modifications are facilitating admixture of wild representatives of the genus Canis with dogs regardless of their evolutionary distance and the environmental conditions. We also found signatures of positive selection for dog-derived gene variants in all three canids. These introgressed variants may facilitate adaptation to humandominated landscapes, particularly by promoting increased reliance on anthropogenic food, which may negatively impact the functional roles of these canids as predators. Our findings imply that the actions required to limit dog introgression in wild canids should include conservation of their natural habitats to maintain low human footprint and reducing free-ranging dog densities in areas with a higher footprint.

# Characterizing the frequency and size distribution patterns of introgressed genomic segments from hybridization in the genus *Canis*

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Natural hybridization may be a positive evolutionary force in some circumstances due to the introduction of beneficial adaptive genetic variation, but due to increasing anthropogenic pressures along with the occurrence of domestic species, their wild progenitors are threatened by introduction of maladaptive alleles, loss of unique adaptations, and genetic swamping. Species representing genus *Canis* (wolves, jackals, and domestic dogs), present an ideal model to investigate hybridization and consequences of introgression of gene variants between them. Genomes of admixed individuals are represented by mosaic ancestry patterns, where the size and frequency of the introgressed segments are influenced by the timing, direction, and rates of gene flow.

We conducted a comprehensive genome-wide analysis to identify the frequency and size distribution patterns of introgressed blocks resulting from hybridization between the *Canis* species across Eurasia. We analysed  $^{\sim}$  200K genome-wide SNPs in wolves (n=315), jackals (n=478), and dogs (n=593). Using multiple analytical approaches, we identified hybrids and estimated ancestry proportions in admixed individuals, excluding F1 hybrids. We then used local ancestry analysis results and a custom-made script to detect number and lengths of ancestry blocks in admixed individuals.

We identified extensive variation in the size and frequency of introgressed genomic blocks among individuals and populations of hybridizing *Canis* species. The sizes of introgressed blocks varied across admixed individuals, ranging from 932 bp (dog-to-jackal) to 94 Mb (wolf-to-jackal). A higher number of introgressed blocks were identified in admixed wolves derived from dogs (1806), followed by jackals derived from wolves (611) and jackals derived from dogs (361). In contrast, fewer jackal-derived introgressed blocks were detected in an admixed wolf (3) and dogs (8). Notably, a genomic segment of 7.72 Mb on chromosome 9 was detected in all eight dogs with jackal ancestry, suggesting potential adaptive introgression. These findings may provide insights into the complex interplay of gene flow and selection in shaping the genomic landscape of hybrid *Canis* populations.

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## Genetic basis of vocal communication in canines: insights from FoxP gene expression

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Domestication has significantly reshaped canine vocal behavior. Unlike wolves, modern dogs bark frequently and with variety. Ancient breeds like the Basenji and New Guinea Singing Dog retain wolf-like vocalizations (e.g., howling, whining), suggesting a gradient of change. While behavioral shifts often reflect gene expression, the molecular basis of canine vocalization remains poorly understood. The Forkhead box P (FoxP) gene family, particularly FoxP1, FoxP2, and FoxP4, plays key roles in neurodevelopment and vocal learning across vertebrates. FoxP2 is essential for human speech, vocal learning in songbirds, and vocal modulation in bats, while FoxP1 and FoxP4 contribute to broader neurodevelopment and brain formation, respectively. We performed Nanopore direct RNA/cDNA sequencing on wolf and dog (Beagle, German Shepherd, Labrador) forebrains, aligned to ROS\_Cfam\_1.0. Of 95 differentially expressed transcripts (62 downregulated, 33 upregulated in wolves), FoxP4 showed ~14-fold lower expression in wolves, suggesting domestication-related plasticity. A FoxP1 isoform (XM 038565865.1) was six-fold reduced in wolves, while Ncor2 (a FoxP corepressor) was ~5,000-fold lower, pointing to a potential mechanism for enhanced FoxP-mediated transcriptional activity in domestic dogs. These results indicate that domestication targeted FoxP4-related pathways, likely facilitating the emergence of barking as a context-flexible and socially salient communicative trait. Concurrently, dogs exhibit FoxP1 isoform-specific regulation and enhanced Ncor2-mediated repression, which may collectively refine vocalization-related neural circuitry. These findings establish a molecular link between FoxP gene networks and the evolution of vocalization in canids, shedding light on regulatory changes during the domestication of humanity's oldest animal companion.

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# Evaluation of the domestication syndrome hypothesis using genotype-phenotype associations in a free-ranging dog population

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Co-variations between phenotypic traits can be explained by selection exerted on different traits during adaptation to a changing environment, or by the selection for one trait constraining other traits. Understanding how the adaptive process is subject to such constraints is of fundamental importance for our understanding of evolutionary mechanisms. The domestication syndrome in mammals is an example of correlated phenotypic changes during adaptation to a human-modified environment, and is defined as a set of similar traits among domesticates that differ from their wild relatives. It has been suggested that the selection for tameness is the driving force behind the suite of behavioral and morphological changes that define the syndrome. However, this hypothesis has poor empirical support and has been challenged in recent studies. Additionally, while a common causal mechanism involving the development of neural crest cells has been proposed, this hypothesis has not been directly evaluated.

To fill this gap, we studied a population of free ranging dogs in Morocco, measuring traits considered to be part of the domestication syndrome (shape of tail and ears, muzzle length, coat color, and several behavioral traits, including measures of tameness), and obtained genome-wide SNP genotypes for 315 individuals. Using a genome-wide association approach, we identified candidate loci associated with each trait. While none of these loci are shared between the traits, some are located within or near genes that are involved in the development of neural crest cells (e.g., MITF). We also used structural equation models to test the hypothesis of a shared causal mechanism for all phenotypes, including all identified loci. Overall, our results do not support a shared direct genetic basis for the domestication syndrome traits based on pleiotropy, and the candidate genes do not support a common functional pathway directly linking the traits. Nevertheless, we found support for a shared causal mechanism for all traits, which could be explained by more complex underlying processes.

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## Invisible passengers, visible impact: microbiome adds to raccoons' invasiveness

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Invasive alien species establish populations outside native range and pose a major threat to native species and ecosystems, being the second leading cause of species extinctions. Understanding factors influencing their adaptive advantage in the invasive range is crucial for mitigating harmful impact.

Microbiome affects hosts fitness and may be a source of plasticity upon encounter novel environmental conditions by increasing tolerance to abiotic stress, stimulating immune response, protecting from pathogens, widening dietary niche and introducing new metabolic pathways. While microbiome associations are known to impact species survival, the extent that microbiome variation aids in invasion dynamics is unknown. In the presented research, we investigated the diversity and composition of gut microbiome in native and invasive raccoon populations, to assess its role in invasion success. We studied three invasive populations (Germany, Poland, Czech Republic) with different demographic background and invasion history and two geographically distant native populations from USA. We found that native and invasive populations possess distinct microbiomes, and invasive populations display greater gut microbiota diversity without signs of instability or functional decline. Significant differences were found between gut microbiota of each invasive populations pair, suggesting acquisition of local microbial taxa during invasion. The core microbiome of introduced populations consisted of more taxa than in native populations, suggesting a high stability in the invasive range. We found no evidence of reduced gut microbiota diversity linked to the population bottleneck, nor increased microbiome dispersion indicating lower community stability. Native populations exhibited no microbiome differentiation between distant locations and lower, but still reasonably high alpha diversity, suggesting the microbiome stability. The relatively low level of taxa's functional differentiation between native and invasive populations and detection of numerous, but not necessarily overlapping components of healthy gut microbiome suggest adaptation to their respective habitats, although through divergent patterns of microbial composition. Our findings imply that invasion changes the gut microbiota composition, while preserving functional capability. Moreover, this study highlights the gut microbiome as a factor facilitating successful adaptation, through different host-microbiome associations.

# Between geography and forest naturalness, which plays a more significant role in shaping the genetic diversity of saproxylic beetles?

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Saproxylic beetles, dependent on deadwood and forest habitats, serve as important indicators of forest ecosystem resilience and biodiversity. Understanding their genetic structure provides insights into historical biogeography and current environmental impacts. This study uses dd-RADseq to generate genome-wide SNP data from multiple saproxylic beetle populations, including rare and common species sampled across Poland's primeval and managed forests along an east-west gradient.

We tested two hypotheses: (1) Whether Poland is a transient zone for genetic variation, with beetle populations in westernmost and easternmost forests showing strong genetic differentiation due to historical refugia and postglacial recolonization; (2) If forest management affects genetic diversity and population structure, with primeval forest populations maintaining higher diversity than those in managed forests.

Analyses revealed geographic genetic structuring with clear east-west differentiation and some admixed intermediate populations, supporting the transient zone concept. Populations from primeval forests exhibited greater genetic diversity and distinct composition compared to managed forests, notably in rarer species. Common species showed similar diversity across habitats, and some pest species displayed opposite patterns.

This work underscores the importance of combining genomic and habitat data to understand evolutionary and ecological processes shaping saproxylic beetle populations and informs conservation strategies to preserve genetic diversity and ecosystem function amid anthropogenic change.

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# Do Core Bumblebee (Bombus) Symbionts Remain Core in the Nordic Cold?

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Bumblebees (Bombus) represent a diverse and ecologically significant group of insects. Their broad distribution across temperate ecosystems and ability to fly under harsh weather conditions make them effective pollinators. Despite their ecological importance, bumblebee species diversity is dramatically declining. The precise causes of these declines are not fully understood, nonetheless, numerous biological and environmental factors have been identified, including climate change, reduction in forage plant diversity, and pathogen spillover. Notably, many of these stressors may also perturb the structure and function of bumblebee-associated microbiota.

Bumblebees are known to harbor a limited, well-defined consortium of host-specific, beneficial bacterial genera that constitute the so-called gut-associated core microbiota. Although recognised as a part of their host adaptation, our understanding of microbiota dynamics is still far from complete. To fill this gap, we analyzed the gut microbiome of 137 bumblebee individuals representing 13 species collected in Sweden and Greenland (Zackenberg Valley). Simultaneous amplicon analysis of the bacterial hypervariable regions V1V2 and V4 of the 16S small ribosomal subunit and cytochrome oxidase subunit I (COI) gene fragments enabled both qualitative and quantitative assessment of microbiota as well as host species identification. In turn, metagenomic analyses provided a comprehensive perspective on microbiota composition and facilitated the reconstruction of functional genes and specific bacterial strains in selected specimens.

Our results showed that one of the key genera of the core symbionts of bumblebees (*Gilliamella*) was lost in those from Greenland, questioning the concept of universally conserved core microbiota. Conditions in arctic environments are harsh, with short seasons, scarce food sources, and temperatures near internal ice formation. Core bumblebees' symbionts may be replaced by environmental bacteria due to either the difficult conditions or the substantial fitness costs imposed on the host by symbiont maintenance. The study aimed to describe the dynamics of the bumblebees' microbiota across harsh northern environments. Our results allow us to better understand the dynamics between hosts and their microbiota in the context of adaptation to a changing environment. The crucial aspect in an ongoing insect apocalypse.

# Environmental protist cell screening provides evidence for recent switch to endosymbiosis in bacterial lineage TC1

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Symbiosis is a potent driving force of evolution that contributes to both the major innovations as well as diverse minor adaptations. Among its variations, endosymbiotic interactions often lead to significant changes in both partners and are now considered to be the underlying processes behind the eukaryogenesis and organelle establishment. Significant part of such relationships involves prokaryotes as endosymbionts, performing various roles ranging from metabolic supplementation to host protection.

However, our understanding of endosymbiosis mechanisms and its impact remains poor. Historically predominant body of research was focused on the endosymbiotic associations with multicellular organisms. Eukaryotic microorganisms (protists), representing the highly diverse majority of eukaryotic lineages and known to participate in numerous associations with prokaryotes were much less studied. In addition, the studies on protists are often hampered by the challenges in their cultivation. Together this prohibited from deeper inquiry into the diverse and potentially ecologically relevant interactions, in addition much closely resembling the setting for the organelle acquisition.

Here we report an identification of the new member of enigmatic Bacillota class TC1 associated with the anaerobic ciliate cell isolated from the anoxic zone of the freshwater lake as a part of environmental protist cell screening efforts. Phylogenetic analyses place it as a sister species to endosymbiont TC1 of *Trimyema compressum* ciliate, with unknown function but reported positive impact on the host cell growth. Using the obtained metagenome-assembled genome of the novel bacterium we compare its predicted metabolic capabilities with that of the TC1 endosymbiont and related lineages. Our analyses show that new TC1 class member exhibits less genome reduction than the *Trimyema compressum* symbiont, but likely shares the similar host-associated lifestyle, contrasting with sister genera and suggesting the recent switch to (endo)symbiotic lifestyle. Our findings may help identify specific genomic changes that contributed to symbiosis establishment in this lineage and underscore the importance of search for novel relatives of established symbionts.

## Methanogenic symbionts of anaerobic ciliates are host and habitat specific

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Anaerobiosis has evolved independently multiple times within the phylum *Ciliophora*. Most anaerobic ciliates have changed mitochondria and shifted to anaerobic metabolism, resulting in the formation of mitochondrion-related organelles (MROs). Additionally, anaerobic ciliates have acquired prokaryotic symbionts to scavenge metabolic end products from the MROs and aid in energy production. Several ciliate lineages represent strictly anaerobic groups within the phylum. Here, we explored the diversity of methanogenic symbionts in members of the diverse, free-living, obligate-anaerobic ciliate order Metopida to detect symbiotic specificity patterns. The survey included 54 strains from 32 species from freshwater and marine/brackish. Three methanogen genera were found: *Methanoregula* and *Methanobacterium* in freshwater hosts and *Methanocorpusculum* in marine/brackish hosts, suggesting a strong host-environment specificity. At the higher taxonomic levels, metopid ciliates also tend to harbor specific methanogen genera. This indicates that the host-methanogen relationship remains stable over short timescales, but metopids tend to replace their methanogens over longer evolutionary periods. Thus, anaerobic ciliates use a mixed transmission mode to maintain and replace their methanogenic symbionts, enabling them to thrive in oxygen-depleted environments.

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# Exploring phenotypic and microbial variation in *Philaenus* spittlebugs

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Auchenorrhyncha – a diverse clade of sap-sucking insects that includes leafhoppers, planthoppers, spittlebugs and cicadas – is a valuable system for symbiosis research. For the majority of its representatives, plant sap is the only source of nutrition, but it is deficient in essential amino acids and other nitrogenous compounds. To overcome this limitation, Auchenorrhyncha established mutualistic relationships with microorganisms. It is estimated that this symbiosis originated approximately 300 million years ago when their last common ancestor was colonised by a bacterium from the phylum Bacteroidetes, now known as Sulcia. In numerous Auchenorrhyncha lineages, Sulcia still persists alongside with co-primary or/and facultative symbionts, forming complex symbiotic systems. This study specifically investigates Philaenus spumarius, a widespread, polymorphic spittlebug species and a major vector of the plant pathogen Xylella fastidiosa. It displays over 16 genetically determined dorsal colour morphs, the frequency of which varies across geographic regions and environmental conditions. While the symbiotic partners of these insects have been partially characterised, the relationship between morphotype diversity and microbiome composition remains poorly understood. Therefore, we collected over 300 individuals from populations across mainland Sweden and Gotland and Öland islands. Among them, based on external patterning, we identified approximately 20 distinct Philaenus morphotypes, 14 of which were exclusive to Gotland, suggesting strong regional differentiation. In order to verify the genetic diversity of examined specimens as well as determine their microbial associates, multitarget amplicon sequencing was performed on all individuals. Our results confirmed the consistent presence of Sulcia, represented by a single genotype across all samples. In contrast, both Sodalis and Wolbachia were widespread but displayed high strainlevel diversity, with distinct variants occurring in different populations. While no clear correlation between colour morph and microbiome composition has yet been observed, further analyses are conducted to explore subtler patterns. To conclude, this study highlights the ecological and evolutionary complexity of intraspecific diversity in P. spumarius and provides new insights into how phenotype, symbiont diversity, and geography may interact in shaping host-microbiome relationships in Auchenorrhyncha.

Kleptoplasts in *Rapaza viridis* reveals early and intermediate stages of plastid integration and the phylogenetic origin of the plastid proteome

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Acquisition of plastids by the fixation and reduction of a photosynthetic endosymbiont requires the host cell to gain new genes in its nuclear genome encoding for proteins then targeted into the plastid. Theoretical evolutionary models predict that the host acquires these genes by massive gene transfer from the endosymbiont. Unexpectedly, phylogenetic studies have reported numerous donors of these proteins, suggesting that horizontal gene transfer from species other than the ultimate endosymbiont can be as significant as those that originated in the endosymbiont. However, the ancient of such events usually precludes precisely detecting the donor of these proteins. The mixotroph Rapaza virdis acquires a temporary plastid by kleptoplasty over a green unicellular alga. Surprisingly, genomic analyses of its protein repertoire detected numerous nucleus-encoded plastid-targeted proteins, unlike most other kleptoplasty examples, suggesting this species as a model representing an early stage of plastid acquisition. In the study, we investigate the phylogenetic origin of each of the more than 200 proteins of the Rapaza viridis plastid proteome, as predicted from transcriptomic data. We complement this dataset with newly obtained transcriptomic data from seven photosynthetic and non-photosynthetic Rapaza viridis relatives, as well as a broad representation of eukaryotic proteins from all known lineages harboring plastids. Reconstruction of the metabolic pathways with proteins predicted to be plastid-targeted revealed an intimate integration of the kleptoplasts and the Rapaza viridis metabolism, including photosynthetic machinery and carbon fixation, as well as the biosynthesis of other compounds, such as chlorophyll and terpenoids. However, most of these pathways were not recovered completely, suggesting an ongoing process of integration. The phylogenetic analyses of these proteins revealed a mosaic origin of the plastid proteome in Rapaza viridis. While a significant proportion of the proteins proceeded from different donors, no single species contributed as much to the plastid proteome individually as the endosymbiont. These results shed light on the early steps of plastid acquisition and the proportion of different actors in this process.

# Condition-dependent expression of hyaluronic acid receptors on spermatozoa: evolutionary implications for cryptic female choice

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Fertilization represents a critical stage of post-copulatory sexual selection in many taxa, during which molecular interactions between gametes influence reproductive success. Reproductive failure, however, has become an increasing concern in modern society. Recent studies suggest that, in addition to pathological conditions, fertilization failure may also result from post-mating sexual selection mechanisms acting on the level of gametes. After mating, interaction between the sexes is limited to biochemical communication between sperms and female reproductive tract secretions, which may facilitate fertilization bias towards specific males, also known as cryptic female choice (CFC). However, the molecular basis of that process remains unclear. Here, we explore the assumptions of condition-dependent model in the context of post-mating sexual selection as a putative mechanism of CFC, driven by female reproductive tract fluids in response to male condition. We measured the RNA expression of two genes coding for crucial fertilization-related hyaluronic acid (HA) receptors, CD44 and HMMR, and correlated this with sperm performance and male condition. Condition-dependent variability in CD44 and HMMR expression on the sperm surface may serve as a cue for the female and be a subject to selection, enabling cryptic choice through preferential fertilization. Understanding the molecular basis of CFC provides insight into the evolutionary interplay between male and female reproductive strategies, with potential applications in improving assisted reproductive technologies by mimicking sexual selection mechanisms.

# Swipe Right for Survival? Mate Choice Symmetry and Extinction Risk.

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Environmental change threatens population persistence, but the role of sexual selection in adaptation is debated. We developed an individual-based simulation model to explore how mate choice systems influence genetic diversity, effective population size, and extinction risk in dynamic environments. The model contrasts mutual mate choice (monogamous to polygynous) with female-only choice to assess how male choice and polygyny affect genetic diversity and population resilience under fluctuating conditions. We investigate whether extra-pair copulations in monogamous mutual choice systems lower extinction risk by enabling mating with high-quality males, thus enhancing adaptation. The model also examines how mate choice and mutation interact to aid genetic resilience against multiple environmental stressors and how sexual selection mitigates genetic drift in populations with limited gene flow. Sexually selected traits are genetically encoded, condition-dependent, and responsive to diverse environmental dynamics (directional, stepwise, random, or stable). By simulating population size, trait evolution, and extinction risk, we hypothesize that asymmetric mate choice amplifies adaptive potential by favoring better-adapted males and extra-pair copulations to increase adaptive potentials in mutual mate choice systems. This framework offers novel insights into how sexual selection symmetry shapes population responses to environmental change.

# Scaling up sexual selection: Microevolutionary processes predict speciation in *Brassicaceae*

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Sexual selection is a recognized driver of trait diversification in animals, but its role in shaping speciation remains contested. In plants, sexual selection operates both before pollination—via pollinator-mediated competition—and after pollination through pollen competition and pistil-selective traits, yet its macroevolutionary consequences are poorly understood. Here, we test the hypothesis that stronger sexual selection promotes higher speciation rates in Brassicaceae by accelerating the evolution of reproductive barriers.

We used petal size (length and width) and pollen-to-ovule (P:O) ratio to represent pre-pollination sexual selection, and style length as a proxy for post-pollination mechanisms. Trait data were obtained from direct measurements, literature, and machine learning imputations. Mating system and ploidy were included to control for confounding factors. Phylogenetic analyses were based on a 1004-species tree, time-calibrated in BEAST2 using nuclear and chloroplast markers. Speciation rates were estimated using ClaDS ( $\lambda$ \_tip), BAMM ( $\lambda$ \_BAMM), and the DR statistic ( $\lambda$ \_DR). We also quantified evolutionary rates and rate shifts for each trait using BAMM and RRPhylo. Associations between traits and speciation were tested using STRAPP and ES-sim. HiSSE models assessed the independent effects of mating system and ploidy on diversification.

We found that the mating system significantly predicted all sexual selection proxies, whereas ploidy had no effect. Selfing lineages diversified more slowly than outcrossing ones. Among the trait proxies, P:O ratio showed the strongest positive association with speciation across all methods, followed by style length. Petal traits showed weak or non-significant correlations. Evolutionary rates and rate shifts for P:O ratio and style length were also elevated in lineages with higher speciation rates.

These results support a role for sexual selection in promoting plant speciation, with pre-pollination mechanisms—especially P:O ratio—emerging as stronger predictors than post-pollination traits. Our study highlights sexual selection as a relevant but underexplored driver of plant diversification. Future work integrating fitness estimates, ecological context, and genomic data will be key to disentangling how sexual selection shapes macroevolutionary patterns in plants."

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## Impact of food quality and quantity on the life history of *Stenostomum* (Platyhelminthes: Catenulida)

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Microscopic, free-living flatworms from the order Catenulida often inhabit temporary freshwater habitats, in which they prey upon unicellular eukaryotes and microinvertebrates. Catenulids reproduce primarily asexually through paratomy, a process in which new head structures are inserted in the trunk of the fully developed worm, followed by the transverse fission that separates the offspring. However, in some species, the regular paratomy might be altered – the new individuals (zooids) are produced, but do not separate. This leads to the development of so-called chains – linear conglomerates of multiple zooids that remain connected tails-to-heads. Here, we tested whether the chain formation can be triggered by different types and concentrations of food in the common catenulid genus, *Stenostomum*. First, we fed six species of *Stenostomum* with seven different prey species (six unicellular eukaryotes and one rotifer) to assess which food is sufficient to maintain asexual reproduction in particular flatworm species. Since the ciliate *Paramecium bursaria* turned out to be the most universal food item, we next tested the impact of its abundance on chain formation in *Stenostomum*. Indeed, high concentrations of *P. bursaria* triggered temporary chain formation in two of the tested species. Therefore, it appears that chain formation is a response to food quantity and not quality. We suggest that chains result from an accelerated cell division rate, which – under excessive food conditions – surpasses the rate of morphogenesis, leading to the establishment of multiple developing heads in a single organism.

### Constrained evolution of woodpecker foraging ecology across the world's environments

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It has long been hypothesized that woodpecker foraging strategies exhibit broad geographic patterns. For instance, wood excavation and trunk foraging are markedly less frequent in tropical than in temperate forests. In contrast, tropical woodpeckers more often engage in probing behaviors, foraging in crevices, epiphytes, or among thin branches. If such patterns are consistent, they may reflect deeper evolutionary processes. Two main hypotheses could explain this variation. First, environmental specificity, whereby forest types differ in wood density, bark thickness, and epiphyte cover, thus shaping foraging behavior. Second, climatic constraints on morphology, whereby species in colder climates evolve larger bodies (Bergmann's rule) and smaller appendages like beaks (Allen's rule). Notably, smaller-bodied and longer-billed species are more often probers adapted to canopy foraging—suggesting that climate may indirectly shape behavioral evolution even when alternative strategies are ecologically viable. In this presentation, I share ongoing results from a global, species-level comparative analysis of woodpeckers, applying phylogenetic methods including path analysis and Ornstein—Uhlenbeck models to evaluate these scenarios.

### Modulation of rest-phase hypothermia and its energy-saving efficiency in ageing zebra finches

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Most birds lower their body temperature during periods of inactivity, a phenomenon known as rest-phase hypothermia. While this response is generally considered an energy-saving strategy due to its associated drop in metabolic rate, its effectiveness may decline with age as thermoregulatory capacity deteriorates. Consequently, the energy balance of older birds may become compromised. This is particularly impactful in energetically-challenging conditions, such as food scarcity, to which birds commonly respond by deepening rest-phase hypothermia. However, this solution may become unattainable for older individuals, limiting their capacity to maintain body mass. Although both fasting and ageing have been studied independently, their combined impact on the avian hypothermic response remains poorly understood. Addressing this gap requires an integrated approach to the physiology of avian hypothermia.

Therefore, we continuously measured body temperature and metabolic rate in zebra finches of different ages. We tested how these physiological parameters contribute to birds' ability to maintain body mass with or without food access. When fasted, older birds lowered their metabolism more than young ones, resulting in lower average and minimal body temperatures. Simultaneously, they took longer to reach minimum body temperature regardless of food access. Despite clear changes in metabolism and thermoregulation, body mass losses were similar in old finches compared to young ones. This may suggest that with age, defending stable body mass requires a progressively deeper rest-phase hypothermia in order to save enough energy, especially when resources are scarce. Data collected with our combined approach allows for studying complex interrelations between age and various aspects of avian physiology.

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### Long-term selection for divergent BMR affects reproductive outcome in mice

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Basal metabolic rate (BMR) is a benchmark metric in biology and biomedical studies. It quantifies the energy metabolism in an adult endotherm at rest, within thermoneutrality, and in a post-absorptive state. Since BMR varies not only between but also within species, one could expect it to have a profound impact and consequences for the individual's performance and fitness, including various trade-offs on energy allocation. Here we tested how long-term selection for divergent levels of BMR applied over 60 generations affects the reproductive outcome and patterns in laboratory mice (High-BMR and Low-BMR lines). We found that litter size diverged between the two line types following the pattern of selection progression, separating significantly c.a. generation 45 onward, with the size of weaned litters in the Low line relatively decreasing at a rate of 0.66% per generation and relatively increasing at rate of 0.15% per generation in the High line. Having a multigenerational track record also allows us to analyze if the selection protocol resulted in any adaptive offspring sex allocation changes or seasonal effects. This showed the sex ratio was overall skewed toward males in the litters, and was not affected by the line type. The season seems to be also a significant factor when it comes to reproduction, with both line types having the smallest litters in winter and the largest in spring and summer.

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### Plasticity of the skeleton in lactating female mice

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Lactation is one of the most energetically costly periods in a female mammal's life. Effective heat dissipation during reproduction seems crucial to prevent hyperthermia during extensive milk production (according to Heat Dissipation Limit theory), especially in the face of global climate warming. Because the small rodent body's most effective parts in dissipating the surplus heat are extremities like the tail or feet, one can expect plasticity of the size of those parts in females during reproduction. Their elongation and increasing surface-to-volume ratio might adapt to rearing bigger litters, especially in animals with high body maintenance costs (Basal Metabolic Rate – BMR). We tested this prediction using mice divergently selected for a low and high level of BMR.

For two groups of females (non-reproductive and post-weaning) from both selection line types, we measured the lengths of the tails (from the back of the sacrum to the end of the tail), spines (from the atlas (C1) vertebra to the sacrum), and sacral bones using radiography (RTG). Lastly, we recorded the number and length of all vertebrae in the tail and lumbar spine to identify the source of the observed differences in tail and spine lengths.

The body mass-corrected size of all measured skeleton parts significantly differed between the line types and reproductive statuses. Moreover, we observed a significant interaction between these factors. We confirmed the hypothesis of a significant correlation between the level of BMR and the size of extremities used for cooling the body. Additionally, increasing energy expenditure (milk production) results in elongation of the female body (measured as spine length) and their tail. The elongation of the spine and tail was the result of the growth of vertebrae in these parts of the skeleton, which suggests a high plasticity of the mouse skeleton not only during the period of the most intensive development of young animals (2 months of their life) but also in adult animals. However, plasticity margins appear to be partly depleted in mice with a high level of BMR.

## Thermoregulatory response to water shortage in aging zebra finches

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In nature, birds can encounter factors challenging their energy balance, such as low ambient temperature, food scarcity or water shortage. A common response to such a challenge is deepening of rest-phase hypothermia, which is assumed to reduce energy expenditure. However, such modulation relies on efficient thermoregulatory mechanisms, which may deteriorate with age. In this study, we investigated how the absence of drinking water affects body temperature regulation in zebra finches (*Taeniopygia guttata*), a species capable of relying solely on metabolic water. Specifically, we examined whether such thermoregulatory changes help conserve body mass and how these processes are influenced by age.

We used zebra finches representing a broad age span and quantified changes in their body mass and circadian body temperature pattern in presence or absence of drinking water. Without access to water, the birds experienced a substantial body mass loss which stabilized within six days, and this decrease was more severe in older birds. Within the first three days, the water-deprived birds deepened their rest-phase hypothermia and lowered their body temperature towards the end of the day. The scope of the response was not affected by age. Overall, water shortage poses a challenge to a zebra finch energy balance, and the birds respond by modulating their circadian temperature pattern. However, efficiency of coping with this challenge appears to decrease with age: despite a similar scope of the response, older birds experienced more severe body mass loss than the younger ones. Hence, aging may increase vulnerability to changes in water availability.

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### Wing Morphology as a Test of Trophic Differentiation in Cryptic Bat Species

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A species is the basic unit of biological diversity, evolving as a single lineage of populations while maintaining its distinctiveness and evolutionary tendencies. This also applies to cryptic species, which are morphologically very similar but genetically distinct. This (apparent) lack of morphological differences, combined with sympatric occurrence, can be seen as an evolutionary phenomenon, because according to Hutchinson's limiting similarity theory, niches can only overlap to a limited extent.

Morphological divergence should be driven by adaptations to different niches, with food acquisition typically being the strongest differentiating factor. Therefore, cryptic species are excellent model organisms for studying Hutchinson's limiting similarity theory — given their overall similarity, even small structural differences may indicate niche specialization.

Bats are a suitable subject for such research because their adaptation to flight limits morphological variation. Also, changes in wing or uropatagium shape are likely to reflect adaptations to specific flight types as adaptation of predator to prey capture strategies.

Three groups of cryptic bat species from three different genera were selected for study: *Myotis alcathoe, M. mystacinus, M. brandtii, Rhinolophus euryale, R. meheleyi, Plecotus auritus, P. austriacus, P. macrobullaris.* These species have well-documented geographic ranges, occur in the same habitats within each group and species composition of their prey is known from molecular studies.

We took standardized photographs of wet specimens and measured each bone lengths to assess wing shape (e.g. proportion of length of 3rd to 5th finger, aspect ratio) and uropatagium shape. Simultaneously, we measured the wing shape (e.g. 1st moment of area) and size parameters of insects (such as proportions of each body parts), based on photographs.

Our preliminary results show interspecific variation in wing shape within each cryptic species group. For example, *M. alcathoe* has the narrowest and longest wing in relation to its body size, which suggests that it hunts its prey exclusively in flight, while *M. brandtii* has the smallest uropatagium in relation to its body size, which may be a possible adaptation to collecting non-flying prey. The most distinguishing feature of the species is the proportions of the finger lengths indicating the shape of the wing.

**ABSTRACT BOOK** 

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### P1. Bacterial diversity in the blood of synanthropic rodents within Poland

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Rodents are small to medium-sized, highly adaptable mammals capable of thriving in human- modified environments. They can rapidly establish large populations in areas where resources are abundant. Notably, rodents are recognized reservoirs of zoonotic pathogens, with approximately 10% of the global rodent population acting as carriers of pathogens of public health importance. To assess the bacterial diversity and abundance in the blood of synanthropic rodents, a total of 312 individuals were live-trapped across diverse habitats in Poland, including rural, urban, and suburban areas. Blood samples from these rodents were subjected to metabarcoding analysis. Of the 312 samples collected, 281 (90.06%) tested positive in an initial polymerase chain reaction (PCR) screening for bacterial pathogens, with Listeria spp. used as a control. Positive samples were subsequently sequenced using Illumina paired-end sequencing technology after the attachment of group-specific metabarcoding adapters. The resulting next-generation sequencing (NGS) data were analyzed using the QIIME2 pipeline to align sequences and assign taxonomic classification to the bacterial communities present. To further investigate the impact of urbanization on pathogen prevalence and host-pathogen associations, statistical analysis will be conducted using the R programming environment. This study offers better insights into the diversity and prevalence of blood-borne bacterial pathogens in synanthropic rodents and identify the potential public health risks associated with human-rodent interactions in anthropogenic environments. The findings aim to inform strategies that reduce human exposure to rodent-borne pathogens in areas of close cohabitation.

### P2. Life history traits of predatory mites impaired by particle pollution

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Human-induced environmental change is increasingly affecting the availability of habitats and the functional integrity of ecological refugia. Among its consequences, particle pollution (particulate matter (PM)), which contains trace metals from road dust and industrial emissions, has emerged as a ubiquitous ecological stressor. These particles accumulate on plant microhabitats, potentially disrupting plant-associated arthropod life histories and ecological interactions.

We studied how PM deposition affects life history and behavioural responses of in *Euseius finlandicus*, a predatory mite widely used in integrated pest management (IPM). The species use leaf-based microhabitats for oviposition, development, protection, making it ecologically relevant model for assessing how pollution-mediated habitat degradation can drive eco-evolutionary responses in beneficial arthropods.

Mite populations were reared on small-leaved linden (*Tilia cordata*) leaves exposed to three artificial PM levels simulating urban gradients: low (urban park), moderate (roadside), and high (industrial). Control leaves remain unpolluted. The PM consisted of road dust containing metals such as As, Ba, Cd, Cr, Cu, Fe, Mn, Ni, Pb, Pt, and Zn, known as contaminants of urban and peri-urban environments.

Our results show that PM significantly affects the life history traits of *E. finlandicus*. Moderate and high levels of pollution reduced daily fecundity by 47% and 70%, and total fecundity by 62% and 77%, respectively. Key demographic parameters (including net reproductive rate, intrinsic rate of increase, and finite rate of increase) decreased significantly under pollution exposure, especially in the high contamination treatment. Pre-oviposition duration increased, while oviposition duration declined and mortality rates by 19%. Behavioural changes included pollutant adhesion to mite bodies and increased escape behaviour (13% of juveniles, 25% of adult females) in polluted environments, suggesting potential stress avoidance.

Our findings highlight that PM pollution disrupt essential life history traits and increasing stress-induced behaviours in beneficial arthropods. Understanding these effects is crucial for maintaining the effectiveness of biological control agents in urban and peri-urban environments, especially under increasing anthropogenic pressure.

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**ABSTRACTS: POSTERS** 

### P3. Impact of Social Information Broadcasting on Forest Fragmentation and Avian Biodiversity

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Habitat fragmentation is nowadays considered to be one of the greatest biodiversity threats. However, there are few studies addressing how the different biodiversity metrics (taxonomic, phylogenetic and functional diversity) react to the habitat fragmentation when comparing one to another, and how the effects of habitat fragmentation could be mediated by the social public information. Our goal was to examine how biodiversity metrics of bird communities in forest patches change with the increasing isolation of those patches, and how those changes could be mediated by manipulating social information. We have inspected over 150 forest patches in Poland. For each bird community inhabiting a given patch, biodiversity metrics were calculated. After that we have conducted a large scale behavioral landscape experiment in those patches, where we have broadcasted a different types of social information (songs of similar species, voices of predators, neutral sounds etc.). Biodiversity metrics reacted in a different way to changing the values of parameters describing stand features and its isolation. Social information proved to be able to mediate the effects of patch size and isolation. Our research helps to understand the importance of individual patches of forest habitats in the farmland for the preservation of biodiversity.

# P4. Bay vs. open sea: unravelling the diversity of meiofauna in the littoral zone of Kuźnica area, Hel Peninsula, Poland

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Meiofauna are small benthic invertebrates inhabiting both marine and freshwater ecosystems. They are usually defined as organisms that can pass through a 500 μm sieve but are retained on a 40 μm sieve. There is little information on southern Baltic littoral zones in the existing literature and the meiofauna of the Polish Baltic coast has not been widely documented to date. Therefore, we conducted a survey of meiofauna in the littoral environments on both sides of Hel Peninsula at the height of Kuźnica. The survey was intended to widen the available documentation of meiofauna from the region. Furthermore, we expected to observe differences in the faunal composition of both sides of the peninsula, which differ substantially in wave exposure, sediment characteristics, and vegetation thickness. The outer side is an active environment with high wave exposure and sandy sediment. The Puck Bay, on the other hand, is characterized by lower wave exposure, slightly more finegrained sediment rich in organic matter, and dense vegetation. Salinity was comparable in water samples taken from both sides of the peninsula (0,5%). Differences were indeed observed with open sea habitat exhibiting greater abundance of Diurodrilus, a genus of orbiniid annelids, and Collembola being recorded only from the Puck Bay. Moreover, acoels (order Acoela) were observed only in the samples taken furthest away from the beach on both sides of the peninsula. Other organisms documented in the survey include representatives of Platyhelminthes (e.g. Proseriata, Kalyptorhynchia), Nematoda, Copepoda, Cladocera, Acari, Annelida, Rotifera, and Gastrotricha. The documentation includes microscope photographs of organisms of most recorded taxa.

**ABSTRACTS: POSTERS** 

# P5. Safeguarding genetic diversity for *Lynx lynx* conservation: the utility of captive-born individuals and genetic monitoring of reintroduced and native populations

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Conservation of endangered species requires a holistic approach that goes beyond general population reinforcement and includes careful, individual-based management of genetic diversity. As a keystone species and top predator, the Eurasian lynx (*Lynx lynx*) plays a pivotal ecological role in maintaining the balance of forest ecosystems across its native range throughout Eurasia. After extinctions of this species at the western margins of its range, several reintroduction and translocation programs were implemented to restore viable populations. Usually small and isolated reintroduced populations are vulnerable to inbreeding, which lowers genetic diversity, elevates homozygosity levels, and compromises the adaptability and overall fitness of individuals. Maintenance of genetic diversity is, thus, an integral component of conservation planning. Microsatellites are suitable markers for determining genetic diversity levels and detecting inbred individuals, thereby providing necessary information for decision-makers. We used 20 microsatellite loci to assess genetic polymorphism and individual inbreeding of captive-born *Lynx lynx* and compared these results to those from wild populations. We thus provide new, individual-based management recommendations for the reinforcement of the reintroduced *Lynx lynx* populations using captive-born individuals. By a proper genetic assessment of captive-born individuals that are highly variable and exhibit low inbreeding values, we facilitate the selection of animals for reintroduction programs in order to increase the chances of restoration programs of this top predator in Europe.

### P6. Contact zone of two cryptic Alpine newt lineages in the southern Carpathians

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The alpine newt (Ichthyosaura alpestris) is widely distributed across Europe. Molecular studies have identified seven evolutionary lineages of this species, along with their respective distributions and hypothetical contact zones. The Western lineage, which includes the subspecies I. a. alpestris, occurs in Central and Western Europe. This lineage exhibits two contact zones with the Northern Balkans and Romanian lineages. The Romanian populations of the southern Carpathians are genetically distinct from the Western lineage and most closely related to Balkan newts assigned to the subspecies I. a. reiseri – this affinity is supported by both mtDNA and nuclear markers. Based on an mtDNA timetree, those lineages diverged several million years ago and are currently regarded as cryptic. Both meet in a contact zone of hitherto unknown extent at the border between the eastern and southern Carpathians. We analyzed nucleotide divergence across entire mitochondrial genomes (mtDNA) of Western and Romanian lineages, and mitotyped 772 individuals from 60 populations using lineage-specific PCR primers. Additionally, we estimated divergence at 447 nuclear genes and focused on 40 diagnostic substitutions between the two lineages to design a targeted Molecular Inversion Probes for genotyping a subset of 673 individuals. The overall mtDNA divergence between the two subspecies was 7.2%, while across 13 mtDNA proteincoding genes ranged between 5.9% (COX3) to 12.3% (ND3). Average nuclear divergence was lower, at 0.6%, but ranged between 0.2% (ETS complex II genes) – 0.8% (ETS complex IV factor genes). We found a steep and narrow transition in mtDNA, with a zone of admixture of ~20 km width located along the Buzău, Zizin and Tărlung valleys. In contrast, we found a broad transition (~150 km) in diagnostic nuclear alleles suggestive of a wide hybrid zone. High levels of genetic admixture were found across most of the sampled populations of the Southern Carpathians, as well as the southern part of the Outer Eastern Carpathians. This suggest lineage fusion after secondary contact and argues against specific or subspecific status for the southern Romanian alpine newt lineage. This highlights the need for more targeted approach to track down genes with restricted gene flow and introgression patterns to explain the discrepancy between mtDNA and nuclear allele transitions across this area.

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## P7. Tracing long-term anthropogenic nitrogen input in different ecosystems using stable isotope ratios of plant biomass

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We measured  $\delta 13C$  and  $\delta 15N$  values of plants from herbarium specimen collected between 1870 and 1930, as well as contemporary specimen, collected from the same area or possible similar ecosystems. The specimen represented dry grassland species and wetland species. We found a decrease in  $\delta 13C$  values of wetland species which exceeds the shift we could expect to be caused by the Suess effect, which indicates changing water regimes. Our results show an increase by 1.29‰ in  $\delta 15N$  in wetland plants, and a decrease by 3.03‰ in dry grassland species. This suggests, that wetlands are mainly impacted by organic fertilizer and sewage discharge into aquatic ecosystems, while grasslands have a strong sign of anthropogenic nitrogen deposition and/or artificial fertilizer. However, we found no increase in total nitrogen content in plant biomass. We found no correlation between the size of the shift in  $\delta 15N$  and the use of artificial fertilizer per hectare in the adjacent area or the cover of agricultural land.

Herbarium specimen are a unique proxy for long-term environmental change. In our case, different nitrogen sources have opposing isotopic footprints. This allows the identification of the dominating source. But in most ecosystems nitrogen input is a mixture of different sources, it seems impossible to calculate the contribution of each source, for example using mixing models. "

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**ABSTRACTS: POSTERS** 

### P8. First insight into the evolution of the genus *Rhyzobius* (Coleoptera: Coccinellidae: Coccidulini)

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Ladybird beetles (Coccinellidae) is one of the best recognizable groups of beetles. Despite numerous studies, their classification and phylogeny still remain unclear. One of the most problematic groups within this family is tribe Coccidulini, which currently includes more than 30 genera. The enigmatic and confusing genus *Rhyzobius* is the largest genus within the tribe Coccidulini. It is widespread in the Old World, but the majority of species occur in Australian region, where it is the most species-rich genus of ladybird beetles.

Before 2010, *Rhyzobius* had received little attention. Most of its species were described mainly in the 19th and the 20th centuries. Recent studies, both morphological and molecular, questioned monophyly of this genus. Szawaryn & Tomaszewska (2020) analyzed 29 *Rhyzobius* species from all major zoogeographic regions and constructed a morphological matrix. As a result, they proposed dividing the genus into four clades. This was an attempt to evaluate internal relationships within the genus; however, its monophyly was weakly supported and relationships with other Coccidulini genera were not tested due to the very limited taxon sampling of outgroups. Moreover, the molecular study by Che et al. (2021) confirmed that *Rhyzobius* is not a monophyletic group, and other Coccidulini genera are intermixed with species of *Rhyzobius*. *Rhyzobius* is also known in fossil record. Species of this genus are known from two amber deposits: two species were described from Eocene Oise amber (~53 Mya) from France (Kirejtshuk & Nel 2012) and three species are known from Baltic amber (~37 Mya) from the Gulf of Gdańsk (Szawaryn & Tomaszewska 2020). Thus, *Rhyzobius* is the most diverse group of ladybird beetles in fossil record.

The aim of this study is to test the monophyly of the genus *Rhyzobius*. As a first step, we constructed a morphological matrix using a broad sampling of *Rhyzobius* species within a broad context of other Coccidulini genera to test both monophyly and generic composition of the tribe. For a better understanding of the evolutionary history of the genus *Rhyzobius*, we also analyzed two new fossil specimens. The studied material included species from entire area of occurrence of taxa belonging to the tribe Coccidulini. Our results show that two newly analyzed fossils belong to two different Coccidulini lineages. Furthermore, our analysis confirmed that *Rhyzobius* is not monophyletic and should be divided into several new taxa.

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### P9. Genetic analysis of Microtus (Terricola) grafi based on mitochondrial ancient DNA

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Within the genus *Microtus* (Arvicolinae, Rodentia), we can distinguish the subgenus *Terricola*. It includes more than 13 species, whose distribution extends between the Atlantic, Caspian, Baltic, and Mediterranean Seas. The most widespread species of the *Terricola* subgenus is the European pine vole [M. (T.) subterraneus]. Owing to the low morphological variation within the genus *Microtus*, unambiguous systematic classification can be a problem. In 1992, in the Late Pleistocene materials from the Bacho Kiro cave (Bulgaria), a distinct species of *Microtus* (*Terricola*) *grafi* was identified and described. Samples taken from the site showed the original morphological features of the first molar (M1) (Brunet-Lecomte et al., 1992).

To resolve the taxonomic position of *M.* (*T.*) *grafi*, we obtained mitochondrial DNA sequences from 19 ancient and 8 modern specimens from the Balkans. We reconstructed the phylogeny based on mitochondrial DNA cytochrome b and included all the *Terricola* species. Samples identified by morphology as *M.* (*T.*) *grafi* were located on the branch formed by the modern specimens of *M.* (*T.*) *subterraneus*. We suggest that individuals previously classified as *M.* (*T.*) *grafi* should be classified within the morphological variation of *M.* (*T.*) *subterraneus* species.

### P10. Rehabilitation and Treatment of Large Predators – Challenges and Best Practices

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The rehabilitation and treatment of large predators such as wolves and lynxes present significant challenges for intervention teams, researchers, veterinarians, and wildlife rehabilitation centers. Capturing these animals for scientific purposes or medical intervention requires a highly experienced team with in-depth knowledge of their ecology and behavior.

Wolves and lynxes follow fundamentally different life strategies, which necessitates the use of distinct capture methods. For territorial wolves, soft-catch traps are employed to safely restrain the animal without causing injury. In contrast, lynxes are typically captured using box traps that allow for secure containment until the response team arrives. The use of alarm systems to signal activity in the trap area is highly recommended. The most effective systems include camera traps equipped with GSM modules that transmit real-time images or videos from the trap site.

A rapid response time is critical. In our practice, the intervention team is expected to arrive within 45 minutes of receiving the alert. Regardless of the capture purpose, animals must be sedated prior to any handling to ensure the safety and comfort of both the team and the animal. The veterinarian is responsible for selecting and administering the appropriate drug and dosage, as well as making decisions regarding subsequent treatment and care.

Rehabilitation of injured or ill predators takes place in specialized wildlife centers equipped to handle these species. Regardless of the duration of the rehabilitation, released animals must be closely monitored. It is recommended to release them with GPS-GSM, satellite, or radio telemetry collars, which allow researchers to respond promptly to any post-release complications. Additionally, camera trap monitoring provides valuable insights into movement patterns and physical condition, including signs of healing or deterioration.

This comprehensive approach to aiding injured predators is complex and resource-intensive, requiring the collaboration of many experts. However, the knowledge gained through such interventions is invaluable for establishing standardized best practices in the care, treatment, and rehabilitation of these challenging yet vital species."

### P11. Shifts in gut microbiota in response to urbanization in a common waterbird

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Urbanization processes in wildlife require adaptive changes at many levels, including, among the others, adaptation to food sources of anthropogenic origin, which may determine the composition of gut flora. Expecting that colonization of the urban environment may induce substantial changes in the gut microbiota, we compared diversity and composition of gut microbial communities between urban and non-urban populations of a common waterbird, the Eurasian coot (Fulica atra). The V3-V4 16S region was sequenced, filtered, and analysed in over 200 faecal samples of Eurasian coots. In total, we identified over 65 thousand amplicon sequence variants (ASVs) representing 55 phyla and 550 bacterial families. We found high alpha diversity of gut microbiota (nearly 1000 ASVs per individual), while taxonomic profiling using the FAPROTAX database identified animal parasites or symbionts as the most dominant group (over 50% of all reads). Most importantly, we found significant effects of urbanization on the gut microbiome in the coot, providing support for urbanization-related enhancement of gut microbial diversity. Our results provide important insights into the influence of urbanization processes on the composition of the gut microbiota in non-passerine birds and they constitute an important starting point for further research on the effects of urban life on avian gut microbiomes.

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## P12. Linking urban ecology and immunogenetics: polymorphism of innate immune receptor (TLR1LB) in an urban bird

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Urbanization profoundly remodels fitness pressures faced by animals living in urban habitats and pathogen threat is often considered among the most important ones. Urban animals must adjust their immune function within the innate and/or acquired branches of the immune system to fight off pathogenic microbes, protect health, and enhance survival. Although urban life may promote immunosuppression due to depauperate diversity of urban pathogens, high population densities of urban-living animals may increase horizontal transmission of microbes, requiring immune adaptation. The feral pigeon Columba livia domestica is possibly the most notorious urban bird and an excellent model to study density-dependent processes in urban environment. Our aim was to investigate immunogenetic variation of feral pigeons in relation to population density and habitat urbanization. We specifically focused on the toll-like receptor (TLR) 1LB gene, which codes an extracellular receptor recognizing molecular patterns of bacterial lipoproteins, thus playing a key role in the innate immune response. To collect data, we sampled 200 feral pigeons in five major cities in Poland, across the gradients of pigeon population density and habitat urbanization. Using Sanger sequencing, we analyzed the single nucleotide polymorphisms (SNPs) within the TLR1LB locus. Overall, we detected 27 unique haplotypes across all the populations, including six haplotypes of >5% frequency. The most frequent (21%) haplotype CoLi\*01 was significantly positively associated with population density, while another haplotype was negatively associated with both population density and habitat urbanization. Our results indicate that population variation of the highly conserved innate immune receptor may be shaped by ecological processes linked with urbanization. Our study suggests that the elevated population density of urban animals may promote genetic-based adaptations within the immune system.

# P13. Use of molecular methods in assessing the distribution, numbers, and persistence of the gray wolf Canis lupus family groups in the Drawa Forest

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The gray wolf Canis lupus is a predator that lives in family groups with exclusive, large territories. The number of this species is increasing in Poland, but monitoring it is challenged by extensive territories and low population densities. In order to obtain information on wolves in the Drawa Forest, a method based on genetic identification of individuals by fingerprinting based on DNA isolated from feces found in the field was used. Eighteen gray wolf individuals were identified from 71 samples obtained from the Drawa National Park after subsequent procedures and analysis. Based on kinship analysis, two family groups and two individuals of undetermined status were identified among the 18 individuals. The family group consisting of 4 individuals was named the Przesieki group. Its territory covers the southern part of the Drawa National Park. A family group of 12 individuals was named the Pustelnia group. Its territory covers the northeastern part of the Drawa National Park. For two individuals, a sufficiently high coefficient of relatedness to the above-mentioned family groups was not obtained, so they were described as individuals of undetermined status. In order to talk about the persistence of gray wolf family groups, the identified modern individuals were compared with 32 archival individuals (2014-2018). A kinship analysis was performed and information about the continuity of the genetic line for the Przesieki group was obtained individuals from this group are related to individuals forming the former Smolarz group, and the territories of the two groups overlap. For this reason, one can speak of continuity of the genetic line for the Przesieki group. For the Pustelnia group and individuals of undetermined status, similar conclusions cannot be drawn. The chosen molecular method provided a lot of valuable information, but in order to draw broader conclusions, studies based on samples collected also outside the boundaries of the Drawa National Park are necessary.

### P14. Microbiome diversity in the phytophagous mite Aceria tosichella

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The microbiome of sap-feeding arthropods plays an essential role in host nutrition, development, and reproduction. These microbes play an important role in the provinsion of nutrients, but the bacterial community can also be influenced by the different host plants on which the herbivores feed. The extent to which symbiotic bacteria contribute to host adaptation in herbivorous arthropods, particularly in specialist versus generalist lineages, remains unclear. *Aceria tosichella* is an obligate phytophagous mite that feeds on cereals (wheat, barley, rye) and transmits plant viruses (Wheat streak mosaic virus, Wheat mosaic virus), and is therefore of economic importance, but without effective population control. The mite has the potential to evolve rapidly in response to environmental conditions. In this study, two lineages of *A. tosichella* were experimentally adapted to constant (one host species, i.e. wheat) and fluctuating (two host species, i.e. wheat and barley) environments. After about 150 generations of experimental evolution, these lineages differ in their host specialisation, being specialist and generalist, respectively. As they differ in the diet consumed, we expect that this may have influenced their microbiome composition. Therefore, we aim to obtain and compare their microbiomes using next-generation sequencing (NGS) and to optimise the method of microbiome extraction. This study will contribute to the knowledge of the potential for niche breadth evolution. It will also help to identify taxa involved in the digestion of plant tissue, which could lead to the development of an effective method of controlling A. tosichella.

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## P15. Condition-dependent mechanisms of gamete-level mate choice (postmating sexual selection) in humans

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Post-mating sexual selection, including cryptic female choice (CFC), may influence which sperm succeed in fertilization1. In humans, recent research suggests that female reproductive fluids (e.g., follicular fluid) can differentially affect sperm from different males2. While studies often focus on HLA genetic compatibility, the functional role of male immune condition in this process remains unclear.

This project tests whether sperm from high-condition males—those with stronger immune systems based on their ability to recognize pathogens via HLA genotyping—perform better in stressful environments and in female reproductive fluids. We hypothesize that sperm from these males are more resistant to DNA damage and maintain higher performance when exposed to follicular fluid.

Fifty to sixty men will be genotyped for HLA and classified into high- and low-condition groups. Sperm from 10 men in each group will be tested under two conditions: (1) exposure to degraded DNA to assess chromatin integrity, and (2) incubation with follicular fluid from 10 women in a full-factorial design. Sperm motility, viability, and DNA fragmentation will be measured over time.

This study will provide new insights into post-mating selection in humans, showing how immune condition—not just genetic compatibility—may shape fertilization outcomes. Results may have implications for understanding human evolution, mate choice, and unexplained infertility, highlighting the complex interplay between immunity, reproduction, and selection at the gametic level.

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# P16. The first direct estimate of the mutation rate in a beetle species from a deep sequenced pedigree of *Ips typographus*

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Accurate estimates of the de novo mutation rate are essential for evolutionary and population genetic inference, yet remain scarce for invertebrates, especially insects. Here, we present the first direct estimate of the single-nucleotide mutation rate in a beetle species, the European spruce bark beetle (Ips typographus), based on high-coverage whole-genome resequencing of a single pedigree (69 offspring). Using a stringent, dual-caller pipeline that includes extensive quality filtering and manual validation of candidates, we identified  $^{\sim}$  50 confirmed de novo mutations, yielding a mutation rate of  $^{\sim}$  4 × 10<sup>-9</sup> mutations/site/generation. Notably, the mutation spectrum was dominated by C>T transitions, consistent with patterns associated with cytosine methylation. However, the limited presence in CpG locations and CpG-rich regions suggests that if methylation is involved, it may follow a non-canonical mechanism. Additionally, we found no significant enrichment or depletion of de novo mutations in either inverted regions or coding sequences, indicating that large-scale genome architecture and functional constraints do not strongly influence the spatial distribution of mutation origins in lps typographus. Our approach follows recent best practices to enhance accuracy and comparability, contributing new empirical data for Coleoptera and expanding the taxonomic breadth of mutation rate estimates.

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## P17. Characterization of repetitive regions in the genome of *lps typographus*

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Repetitive elements, particularly transposable elements (TEs), play a significant role in shaping genome architecture and influencing evolutionary processes. This study focuses on characterizing the composition and genomic distribution of repetitive sequences, with an emphasis on transposable elements, in the genome of the spruce bark beetle (Ips typographus). Utilizing a newly assembled chromosome-level reference genome, the content of repetitive elements was annotated using the Earl Grey pipeline, followed by manual curation and further analysis. Repetitive sequences constitute approximately 51.4% of the genome, with TEs accounting for nearly 23%. The most abundant TE groups identified include long terminal repeats (LTRs), long interspersed nuclear elements (LINEs), and DNA transposons.

A primary aim of the study was to explore the relationship between transposable elements and chromosomal inversions, which are prominent features in the I. typographus genome, comprising nearly 30% of its total size. Two hypotheses were tested: (1) that TE density is elevated near inversion breakpoints and (2) that TEs accumulate within inversions due to suppressed recombination. Contrary to these expectations, transposable elements were found to be significantly depleted within inversions and, to a lesser extent, at their breakpoints. In contrast, gene density was found to be higher inside inversions, suggesting that purifying selection may act to reduce TE accumulation in gene-rich regions to prevent deleterious effects.

These findings highlight the complexity of TE dynamics and their distribution in structurally diverse genomes. Overall, this study contributes to our understanding of genome evolution in I. typographus and provides a foundational framework for future comparative genomic studies within the Curculionidae family.

### P18. Age-specific physiological stress levels in a long-lived seabird, the common tern (Sterna hirundo)

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Birds, characterized by their long lifespan and diverse reproductive strategies, serve as valuable model organisms in gerontological and ecophysiological research. Among them, the common tern (Sterna hirundo), a long-lived, colonial breeding seabird, provides a suitable system for examining how age influences physiological health under natural conditions, and whether any pattern would show sex-specificity. In birds, physiological stress is commonly assessed using the ratio of heterophils (H) to lymphocytes (L), two major types of white blood cells, with an elevated H/L ratio being indicative of a stronger stress response, reflecting the activation of innate immunity and changes in leukocyte profiles. We investigated whether variation in the H/L ratio of common terns was explained by sex and age. Hereto, during the breeding seasons of 2019 to 2024, we obtained 817 blood smears from 485 common terns breeding in a colony located on the North sea coast of Germany. Each smear was stained according to the May-Grünwald-Giemsa method and examined at 1.000x magnification by a single researcher to establish H/L ratios. At the population level, we observed older birds to have a higher H/L than younger birds. This pattern, however, could not be explained by within-individual increases in H/L ratios as birds grew older, suggesting that birds with low H/L ratios selectively disappeared from the population. Age-effects did not differ between males and females and there was no main sex difference in H/L ratio either. Our results highlight the importance of distinguishing within- and among-individual processes in studies of aging and suggest a stress response to be associated with a long lifespan in a wild bird, leading to the question why this is so.

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## P19. The effect of sex on spontaneous physical activity in laboratory mice with different levels of basal metabolic rate

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Spontaneous physical activity (SPA) represents an important component of energy expenditure in laboratory mice. SPA can be affected by several factors. SPA is positively correlated with basal metabolic rate (BMR). SPA can be elevated in lower ambient temperature, when heat generated by SPA can be substituted for thermogenesis and the relative cost of SPA can be reduced. Low SPA is also a risk factor for the development of obesity. Recently, sex has also been recognized as an important but usually overlooked variable that can affect animal's physiological and behavioural traits. Indeed, sex can significantly modulate both thermal biology and SPA in laboratory rodents. Here, we compared SPA in laboratory mice from two lines divergently selected towards either high (H-BMR) or low (L-BMR) level of BMR and acclimated for 30 days to either 23 °C or 30 °C (i.e. thermoneutral zone). SPA was higher in the H-BMR line than in the L-BMR line and in mice acclimated to 23 °C than in mice acclimated to 30 °C. However, sex had no significant effect on SPA. Similarly, there were no significant interactions between the effects of sex, BMR level and temperature acclimation. Finally, there was no clear link between SPA and body mass change during the course of experiment. We conclude that the effect of large difference (>60%) in BMR on SPA did not differ between males and females in studied lines of laboratory mice.

## P20. Artificial light at night differentiates vertical and horizontal distribution of zooplankton at population and community levels

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Artificial light at night (ALAN) affects the distribution of many groups of terrestrial and aquatic organisms, but its effect, especially its range, on zooplankton distribution are poorly understood. In May and June 2020, during the new moon, we studied the vertical distribution of zooplankton in Lake Roś (Masurian Lake District) at distances of 5, 15 and 30 meters from a high pressure sodium (HPS) lamp, placed on a boat, 3 meters above the water surface. The light intensity at the surface was 19.2; 2.8 and 0.7 lux, respectively. Fish distribution was estimated using sonar. The presence of ALAN significantly increased the mean vertical distribution of Bosmina longirostris, Daphnia longispina and Chaoborus flavicans. The effect was observed up to a distance of 15 meters. A much weaker one was observed for Daphnia cucullata, while no significant changes were found for Eubosmina thersites and Leptodora kindtii. The effect of ALAN was stronger in June, when the zooplankton assemblage was dominated by the larger cladoceran species. This effect coincided with a decrease in chlorophyll concentration in the upper two meters of the water column, which may have promoted deeper light penetration and increased the visibility of zooplankton to planktivorous fish. A concentration of fish was also observed in the zone of highest light intensity, where their abundance exceeded 100 individuals/m³. Under natural conditions, the distribution of fish was uniform. The results indicate that ALAN can affect the structure of zooplankton communities and disturb the trophic balance in the lake's subpelagial zone.

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### P21. Al in Evolutionary Ecology Homework: Used, Effective, and Disclosed?

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Unanimous access to artificial intelligence (AI) tools is an opportunity for ease of work and creativity. Researchers and students use them. Are students using them effectively? Are they ready to disclose their use? So far, the first of those questions has not been addressed in the case of university students.

Here, I used data from homework assignments in the Evolutionary Ecology course taught to MSc students over five years (2020-21 and 2023-2025), with the last three years characterized by the availability of AI tools. In 2025, an optional declaration of AI use was added to the assignments. In each year there were between 10 and 31 students, and here I have analysed over 250 of their assignments. In subsequent years, some homework assignments had the same content but different numerical values. In each case, the specific aspect of the homework can be rated as correct/incorrect. These are 1. estimating the population size required to detect inbreeding, 2. calculating the inclusive fitness of individuals helping their kin to reproduce, 3. determining the proportion of individuals at which the population could reach the mean maximum fitness, 4. assessing the interaction term in a 2x2 factor simulation of the persistence of the metapopulation system.

I found that the probability of correct answers was higher in two tasks and lower in two others before and after the introduction of AI tools. The reported use of AI tools varied widely across the four assignments, indicating that students do not use AI ""by default"" but rather when they feel the need. However, at the individual level, there was no relationship between correct answers and reported use of AI.

These results may indicate that students did not understand the nature of the questions or were unable to effectively prompt the AI for correct answers. It is less likely that the tasks were inherently resistant to AI assistance. Either way, these findings may be useful for other teachers of evolutionary ecology. Irrespective of the merits of the tasks, I was pleased to see that the majority of the students, when given the opportunity, filled in the questionnaire regardless of whether they reported using AI or not. This confirms previous findings that students are willing to use AI in an open and professional manner.

### P22. Does the evolution for predatory aggression alter conspecific aggression?

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Aggression, in its various forms, evolved through natural selection to address challenges like resource acquisition and threat defense. Whether predatory and conspecific aggression are linked remains debated. Studying this relationship could shed light on abnormal human aggression, but suitable animal models are limited. We used a unique experimental evolution model: lines of bank voles (Clethrionomys = Myodes glareolus) selected to hunt crickets (Predatory lines) and random-bred Control lines. From generation 36, five males and five females were sampled from each of the four Predatory and four Control lines (total 80). Hunting aggression was assessed before and after mating. On days 12 and 14 post-mating, focal males were placed with a naïve Control male in a neutral arena. For maternal aggression, Control-line intruder males were introduced to mothers' cages on days 12 and 14 post-parturition. All 20-minute encounters were videotaped and analyzed for latency, duration, and frequency of aggressive and defensive behaviors. Voles from the Predatory lines showed greater prey capture success than voles from the Control lines. In intermale aggression test, Predatory males attacked the opponent male quickly, more frequently and for longer duration, while Control males exhibited higher vigilance by spending more time observing the opponent. Among Predatory males, boxing was the most frequent form of attack during the encounters. In the maternal aggression test, Predatory mothers had higher chasing and boxing behaviour than Control mothers, although the effect was not statistically significant. The Predatory voles explored the cage for longer time than Control voles, an indication of their proactive/bolder behaviour which supports conspecific aggression. Thus, selection for predatory aggression elevated conspecific aggression, and this effect was stronger for intermale aggression, suggesting a common genetic basis and the possibility of shared hormonal and neural mechanisms.

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### P23. Molecular dietary analyses of black grouse (Lyrurus tetrix) from two Polish populations

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The black grouse (Lyrurus tetrix) is a species closely tied to a specific habitat type. Thus, the relatively small area occupied, compared to eurytopic species, has to support the complete spectrum of ecological needs: from breeding and brood care, through refuge from predators and sites for leks and territorial behaviour, to food resources. One may therefore expect that the occurrence of such species is strongly shaped by diet and food availability. Understanding how habitat structure shapes diet is key to conserving declining black grouse. We used DNA metabarcoding of fecal samples to characterize plant diets of two Polish populations inhabiting contrasting habitats, the Tatra Mountains and the Biebrza Valley, across phenological periods (mating, breeding, non-breeding). We detected 52 plant taxa spanning shrubs, dwarf shrubs, trees and forbs. Dietary richness per sample was comparable between regions, but composition differed markedly between locations and seasonality. Tatras diets were dominated by dwarf shrubs, especially *Vaccinium sp.*, throughout the year, whereas Biebrza diets showed higher contributions of herbaceous plants and trees. Our results show that black grouse diets track habitat structure and seasonal resource availability rather than differing in overall richness. Conservation should prioritize maintaining habitat heterogeneity that sustains key food resources, bilberry heaths and alpine grasslands in Tatry Mountains, and wet meadows and peatlands in Biebrza, while limiting succession and safeguarding hydrology in lowland wetlands.

### P24. DRB1 gene expression in sperm cells as a potential fertility marker

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One in six individuals globally experiences infertility, and in nearly 30% of cases, the cause remains unknown—referred to as idiopathic infertility. Despite increasing infertility rates and ongoing advances in assisted reproductive technologies, the molecular basis of fertilization is still poorly understood.

Cryptic female choice (CFC) is considered a potential evolutionary mechanism of post-copulatory sexual selection that acts at the level of gametes and may help explain some cases of idiopathic infertility. The recent studies suggests that molecular biocompatibility between gametes is essential for successful oocyte fertilization. One group of genes that may play a role in shaping this compatibility is the major histocompatibility complex (MHC). Within this group, DRB1, a highly polymorphic class II gene, has been proposed as a potential mediator of sperm-oocyte molecular interactions.

We assume that CFC relies on a form of molecular communication between sperm and the female reproductive tract environment. However, no scientific consensus has yet been reached regarding the expression or protein presence of MHC molecules in human spermatozoa. In this study, we aim to investigate whether DRB1 mRNA transcripts are present in sperm cells and whether their expression may influence sperm performance in the presence of follicular fluid.

Although the study is ongoing, our preliminary observations provide initial insight into potential mechanisms of gamete-mediated mate choice. This project represents an early step toward understanding the role of DRB1 in human fertilization and lays the foundation for future analyses of MHC class I and II molecules in the context of CFC.

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# P25. Comparison of the genetic structures of *Borrelia* virulence factor, the outer surface protein C, and the putatively neutral intergenic spacer

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Borrelia afzelii, a major causative agent of Lyme borreliosis in Europe, is known for its high polymorphism of outer surface protein C (ospC) genes that help it evade the host immune system. The OspC protein is essential in the early stages of mammalian infection and is one of the first antigens recognised by the vertebrate immune system. Consequently, its polymorphism has been hypothesised to be due to balancing selection imposed by host immunity, which predicts that it should show a less pronounced genetic population structure compared to neutrally evolving genomic regions.

Here, we tested this prediction by contrasting among population differentiation of *ospC* genes with that of the rrs-rrl intergenic spacer region widely used as a phylogenetic marker characterized by a relatively high neutral rate of evolution.

Analysis of molecular variance (AMOVA) revealed that differentiation between populations explained only 1.84% of the total variance at the OspC, compared to 6.43% variance explained for rrs-rrl marker. The mean pairwise F<sub>ST</sub> values for the intergenic region rrs-rrl ( $^{\sim}$  0.066) region were significantly higher than those for ospC ( $^{\sim}$  0), suggesting a stronger population structure for this neutral marker. The Mantel test revealed a moderate but significant correlation between genetic differences and geographic distance for the rrs-rrl region (r = 0.3624;), while for ospC, this correlation (r = 0.1958) was not statistically significant.

These findings are consistent with the hypothesis that ospC is subject to balancing selection, leading to the maintenance of multiple allelic variants within populations across geographic range. "

## P26. Uneven resources, uneven fish: zooplankton patchiness drives variability in fish growth and metabolism

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Fine-scale spatial heterogeneity is a defining yet underexplored feature of aquatic ecosystems, where prey commonly occur in transient, unevenly distributed patches. Theory predicts that such patchiness influences both average consumer performance and inter- individual trait variation; however, direct experimental tests are limited. We investigated how spatial prey distribution affects physiology and behavior in juvenile rudd (*Scardinius erythrophthalmus*) reared for 40 days under homogeneous or patchy zooplankton conditions, controlling for initial prey quantity. Fish exposed to patchy prey exhibited elevated standard metabolic rate (SMR) and increased foraging effort, but no growth rate (GR) improvement, suggesting energetic costs of prey detection and access offset benefits. Crucially, patchy conditions also increased inter-individual variability in SMR and GR, supporting the hypothesis that environmental complexity drives trait divergence through physiology- environment alignment. Our results show that spatial heterogeneity shapes not only mean trait values but also the distribution of energetic strategies within populations. Such variability may enhance ecological resilience via diversified individual responses but could also amplify selection pressures and disrupt cohort synchrony in persistently patchy habitats. By revealing how prey structure governs physiological performance and trait variability during early ontogeny, our study underscores the ecological and evolutionary importance of fine-scale resource landscapes in freshwater consumer dynamics

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