

Latvian  
State Forest  
Research  
Institute  
“Silava”

12<sup>th</sup> Baltic Theriological Conference  
Baltic mammals in the turbulence of  
anthropogenic tensions

# ABSTRACTS BOOK





# 12<sup>th</sup> BALTIC THERIOLOGICAL CONFERENCE

Baltic mammals in the turbulence of  
anthropogenic tensions

## **ABSTRACTS BOOK**

March 27–29, 2025  
Jaundome, Latvia



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## Abstracts Book

### 12<sup>TH</sup> BALTIC THERIOLOGICAL CONFERENCE

#### Baltic mammals in the turbulence of anthropogenic tensions

Organised by: Latvian State Forest Research Institute “Silava”

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## PROGRAMME

### March 26, 2025

17:00 Arrival and ice-breaking dinner (18:00–20:00)

### March 27, 2025

8:00 Morning coffee & Registration

9:00 Opening

9:20 Plenary speech:

*Marta De Barba, PhD, University of Ljubljana*

A call for sustainability in the use of molecular tools and data for mammal conservation

10:20 Coffee break

10:40 **Baltic fauna news**

*Chaired by Egle Tammeleht*

*Valdis Pilāts*

The cetacean fauna of the Latvian coast including new insights from digitised historical newspapers

*Uudo Timm, presented by Linas Balčiauskas*

Shifts in Baltic mammal populations

*Guna Bagrađe*

Brown bear in Latvia – how long is the road to favorable conservation status?

11:50 **Mammal ecology**

*Chaired by Gunārs Pētersons*

*Jurgis Šuba*

Time series of grey wolf *Canis lupus* and Eurasian lynx *Lynx lynx* Latvian sub-populations exhibit consistency with intraspecific contest competition models

*Christel Rose Bachmann*

Bat habitat use in urban environment based on citizen science data

*Oliver Kalda*

Bat migration over the Gulf of Riga

13:00 Lunch break

14:00 **Molecular methodologies**

*Chaired by Maris Hindrikson*

*Jaana Kekkonen*

Genetic diversity and phylogeography of the endangered Siberian flying squirrel (*Pteromys volans*) over its range

Angelika Kiebler Population genetics of the Siberian flying squirrel in Finland is shaped by glacial retreat and land uplift during the Holocene

Gerrit Wehrenberg Potential role of genetically informed ex-situ conservation for the Siberian flying squirrel (*Pteromys volans*)

Angelika Kiebler, presented by Stefan Prost Gliding towards an uncertain future: opportunities of genomic tools for the conservation of Siberian flying squirrels (*Pteromys volans*)

15:30 Coffee break

15:50 **Molecular methodologies**

Chaired by Laima Baltrūnaitė

Romualdas Lapickis Genetic characteristics of European bison (*Bison bonasus*)

Kirke Raidmets Identifying wolf-dog hybrids in Estonian wolf population with a SNP panel

Kristiina Amur Population genetics of the grey wolf (*Canis lupus*) and the influencing factors in Estonia

Agrita Žunna What can we learn from genetic material obtained from hunted grey wolves in Latvia?

17:30 **Workshop – Large Carnivores** – Hall 1

Facilitated by Peep Männil

18:30 **Workshop – Wild boar & ASF** – Hall 2

Facilitated by Edvīns Oļševskis

19:30 Dinner

## March 28, 2025

8:30 Morning coffee

8:50 Poster session – short presentations

Chaired by Jurgis Šuba

9:30 Plenary speech:

Prof. Arie Trouwborst, Tilburg University

The big picture: ambition in law and policy for mammal conservation and restoration in Europe

10:30 **Wildlife caused conflicts**

Chaired by Jānis Ozoliņš

- Linus Balčiauskas* Did Lithuania experience the anthropause in terms of animal roadkill dynamics?
- Maris Hindrikson* Attitudes towards large carnivores in Baltics: switch from post-Soviet rural into modern urban society
- Petras Adeikis* Wolf predation on livestock management insights in Lithuania
- 11:40 Coffee break
- 12:00 **Wildlife caused conflicts**  
*Chaired by Linus Balčiauskas*
- Mindaugas Bakševičius* Feast or famine: navigating herbivore impacts on regeneration in boreal pine forests
- Gundega Done* Monitoring of deer damage to young pine, spruce and aspen stands, 2018–2023. Data overview
- Gundega Done* Can the presence and structure of undergrowth affect the risk of deer damage to young pines?
- 13:10 Poster session
- 14:00 Lunch break
- 15:00 **Diseases and parasites in mammals**  
*Chaired by Guna Bagrade*
- Jānis Ozoliņš* Wanted dead or alive – wild boar *Sus scrofa* control challenges concerning African swine fever outbreak in Latvia
- Algimantas Paulauskas* Role of mammals in transmission of vector-borne parasites and pathogens
- Marlen Laanep* Parasitofauna of the brown bear (*Ursus arctos*) in Estonia and its seasonal dynamics
- Giedrius Sidlauskas* *Toxoplasma gondii* parasites in the Eurasian beaver (*Castor fiber*) in Lithuania
- 16:30 Conclusions
- 16:50 Coffee break
- 17:10 **Workshop – Power plants & bats & large mammals**  
*Facilitated by Gunārs Pētersons & Jānis Ozoliņš*
- 19:00 Farewell dinner

## March 29, 2025

9:00–15:00 Tour to Razna National Park / Departure

# THE CETACEAN FAUNA OF THE LATVIAN COAST INCLUDING NEW INSIGHTS FROM DIGITISED HISTORICAL NEWSPAPERS

Valdis Pilāts<sup>1</sup>, Carl Chr. Kinze<sup>2</sup>

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*Keywords: porpoise, dolphin, whale, citizen science*

Compilations of cetacean occurrences in the entire Baltic Sea are done by several authors with Latvian records based on museum specimens and scientific reviews. Detailed information on the only indigenous species, the harbour porpoise (*Phocoena phocoena*), was not included or focal to those studies.

Here we employ a new source of information, which has become available through digitized searchable historical newspapers and present our detailed findings on both the harbour porpoise and rarer visiting cetacean species. A total of seven cetacean species have been recorded in the coastal waters of Latvia. Two of them were “discovered” in digitised historical newspapers. Unfortunately, the exact species cannot always be determined from the newspaper texts as animals are usually named simply as dolphins or whales.

Newspaper quotes on the occurrence of the harbour porpoise appear as early as in 1835. The first four decades of the 20<sup>th</sup> century include 25 years with newspaper reports on harbour porpoises whilst the last five decades – only 5 years. Most probably this reflects the overall decrease of the Baltic population. There are at least six newspaper records which could be associated with the bottlenose dolphin (*Tursiops truncatus*). Of these, the earliest record goes back to 1782, but all additional reports are from the beginning of the 20<sup>th</sup> century. The white-beaked dolphin (*Lagenorhynchus albirostris*) was identified from a newspaper record reporting a killer whale (*Orcinus orca*) that had been captured in September 1836. The white-beaked dolphin as a species was only described in 1846 which may explain this misidentification. The next records of dolphins whose description fit with the white-beaked dolphin appeared in newspapers 90 and 100 years later. The common dolphin (*Delphinus delphis*) has been recorded only once in 1853.

The digitised Chronicle of Livonia by Balthasar Russow includes a reference to a whale stranded at the coast of western Latvia in May 1578. The reported size suggests that it was a humpback whale (*Megaptera novaeangliae*) constituting the earliest known record of the species in the Baltic Sea. Newspaper reports revealed two additional plausible cases of humpback whale occurrences in Latvian waters (1931 and 1938). There is no historical evidence of the beluga whale (*Delphinapterus leucas*) and the fin whale (*Balaenoptera physalus*) in coastal waters of Latvia, but both have been recorded relatively recently, respectively in 1982 and 1976.

The authors of newspaper notes on cetacean observations must be regarded as pioneers of citizen science.

## SHIFTS IN BALTIC MAMMAL POPULATIONS

Uudo Timm <sup>1</sup>, Valdis Pilāts <sup>2</sup>, **Linas Balčiauskas** <sup>3</sup>

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*Keywords: mammal declines and extinctions, invasive species, conservation achievements, taxonomy and re-identifications*

Although the total area of the three Baltic countries is relatively small, the differences in the faunal composition of each country are noticeable. There are species with range limits in the region, e.g. *Apodemus sylvaticus*, *Barbastella barbastellus*, *Bison bonasus*, *Glis glis*, *Dryomys nitedula*, *Nyctalus leisleri*, *Pteromys volans*, *Pusa hispida*, *Sorex caecutiens*.

After regaining independence in 1991, the Baltic States underwent significant changes in land use (land reform restoring private ownership and small estates, conversion of military areas into protected areas, changes in agriculture encouraging land abandonment, reduced fertilizer and pesticide use, changes in animal husbandry). The other significant factors were forest dynamics, urbanization, transport, climate change and anthropogenesis. Countries experienced differences in the development of theriology (number of scientists, institutions. and directions), resulting in a variety of long-term and synthesis projects (Red Data and Red Books, studies on invasive species, roadkill analyses, mammal monitoring, population replenishment through breeding, studies on mammalian pathogens, etc.).

In the presentation on changes in the mammal fauna during the first quarter of the 21<sup>st</sup> century, we will cover species declines and extinctions of *Dryomys nitedula*, *Eliomys quercinus*, *Muscardinus avellanarius*, *Pteromys volans*, *Pusa hispida*, *Mustela lutreola*, and discuss the factors influencing these changes. We will also cover new species in the region, such as *Cervus nippon*, *Dama dama*, *Canis aureus*, *Procyon lotor*, *Myopus schisticolor*, *Myotis mystacinus*, *Myotis myotis*, *Neomys milleri*. Changes in taxonomy (*Neomys anomalus* to *N. milleri*, *Erinaceus concolor* to *E. roumanicus*, *Pipistrellus pipistrellus* to *P. pygmaeus*) and re-identifications (*Apodemus sylvaticus* to *A. uralensis*, *Microtus arvalis* to *M. rossiaemeridionalis*) will also be discussed as part of these shifts.

We will present and discuss: (i) the increase in wildlife, especially ungulates, with roadkill of some species increasing exponentially, (ii) initiatives and successes in protecting endangered species and restoring habitats (establishment of *M. lutreola* populations in Hiiumaa and Saaremaa, protection of *P. volans* habitats halting rapid decline, increase in *Lynx lynx*, *Ursus arctos* and other carnivore numbers), (iii) achievements of conservation action plans and regulated hunting, and (iv) the situation with invasive species in the region.

## **BROWN BEAR IN LATVIA – HOW LONG IS THE ROAD TO FAVOURABLE CONSERVATION STATUS?**

**Guna Bagrade**<sup>1</sup>, **Gundega Done**<sup>1</sup>, **Baiba Krivmane**<sup>1</sup>, **Aivars Ornicāns**<sup>1</sup>,  
**Jānis Ozoliņš**<sup>1</sup>, **Digna Pilāte**<sup>1</sup>, **Dainis Edgars Ruņģis**<sup>1</sup>, **Alda Stepanova**<sup>1</sup>,  
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*Keywords: brown bear, Baltic population, large carnivore conservation, monitoring*

The brown bear is classified as an endangered species at the European level, whose protection in Latvia is provided for by Annexes II and IV of the Directive 92/43/EEC. The Action Plan for Brown Bear Conservation aims to promote the favorable status of the Baltic brown bear population in the territory of Latvia over an unlimited time without setting an exact achievement date, or minimum or maximum number of bears. It also aims to ensure the restoration of their range through natural dispersal and the presence of the species as a united and functional component of the wildlife community in man-made and managed landscapes, respecting and promoting the quality of life, well-being and diverse societal interests. According to the “Key actions for large carnivore populations in Europe”, one of the priority measures to be implemented in the Baltic bear population is the expansion of the population range to the south, including the identification of transboundary dispersal routes, comparison of habitat suitability and community tolerance between Estonia and Latvia, and reduction of hunting effort along the southern border of Estonia.

Since 2015, bear monitoring has been carried out with the aim of obtaining data about the status of the population and the habitats required by the species in Latvia. Since 2018, non-invasive sampling has been used to collect material for molecular genetic analyses resulting in 83 individuals genetically identified that have been present in territory between 2018 and 2023. The minimum estimated number of bears in Latvia is 130 individuals (2023 monitoring data). The results confirm an increase in the number of bears, as well as an increase in local population density in the Northern and Eastern regions of Latvia. This increase can be explained both by the arrival of animals from Estonia and other bordering countries to the east, as well as the proven reproduction of bears in the territory of Latvia. In Estonia, the permanent bear distribution range covers almost all of the mainland. The population size has nearly doubled during the last 20 years and is estimated to be 900–950 bears. Bears in Estonia are in favorable conservation status and (mainly due to that) the Baltic population is in the category of “Least Concern” following the IUCN red list assessment. There are no apparent distribution barriers between Estonia and Latvia.

To continue data collection, background monitoring and molecular genetic monitoring should be continued, with particular attention to implementation of genetic monitoring in the border area between Latvia and Estonia.

# TIME SERIES OF GREY WOLF *Canis lupus* AND EURASIAN LYNX *Lynx lynx* LATVIAN SUB-POPULATIONS EXHIBIT CONSISTENCY WITH INTRASPECIFIC CONTEST COMPETITION MODELS

**Jurģis Šuba**<sup>1</sup>, Yukichika Kawata<sup>2</sup>, Andreas Lindén<sup>3</sup>

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*Keywords: wolf, lynx, intraspecific competition, population dynamics*

Two extremes of intraspecific competition – scramble (SC) and contest (CC) – are generally distinguished. The scramble competition involves random resource partitioning among all the individuals in the population. In case of overcrowding, it implies eventual population decline as the available resources per individual decreases. On the other hand, in the contest competition, individuals compete for resources (e.g., sites or refuges), ultimately consumed only by the contest winners as the losers are killed or driven away. Under such circumstances, the winners use as many resources as they need. Frequently applied single-species models of population dynamics, such as Ricker (1954), Skellam (1951), Hassell (1975), and Beverton and Holt’s (1957), are derived from specific assumptions on the type of intraspecific competition and random (R) or aggregated (A) individual distribution.

In this study, we investigated how different population models fitted the available time series of wolf *Canis lupus* and lynx *Lynx lynx* in Latvia, which spanned over six decades. The applied models of population growth, the Ricker (SCR), Skellam (CCR), Hassell (SCA), and Beverton-Holt’s (CCA), were all parameterised in terms of intrinsic growth rate and carrying capacity. A temporal trend was allowed in the projected carrying capacity, justified by an observed increase in prey abundance in recent decades.

For both species, the models of contest competition outperformed the scramble competition models, and the Beverton-Holt model had the greatest weight. However, for the lynx, the difference in performance between the scramble and contest competition models was considerably smaller than that for the wolves. This study promotes the application of population models that reflect intraspecific competition for modelling population dynamics in a single- or multi-species framework.

## BAT HABITAT USE IN URBAN ENVIRONMENT BASED ON CITIZEN SCIENCE DATA

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*Keywords: bats, citizen science, habitat model, urban ecology, habitat restoration*

Urbanization presents significant challenges for wildlife, particularly bats, as their habitats are increasingly modified by human development. Understanding how bats use urban spaces is crucial for effective conservation of this sentinel species. This study utilized a citizen science approach to map bat species, habitats, and activity patterns in an Estonian city, Tartu. Twenty volunteers were trained and surveyed 192 locations, recording bat activity for five minutes per site, with species identifications confirmed by a professional. Across these locations, eight bat species were detected, including *Eptesicus nilssonii*, the Northern bat, which was the most abundant. This adaptable species was found in a variety of habitats, from riverside areas to highly developed urban zones, and was analyzed separately due to its ability to thrive in urban environments. Tall vegetation emerged as a primary predictor of suitable habitats across species, while proximity to water was particularly significant for species other than the Northern bat. Relative air humidity was also found to influence bat activity levels in both species groups, emphasizing its impact on bat behavior within urban environments. This study enhances our understanding of bat adaptation to urban environments and provides valuable insights for conservation strategies aimed at mitigating the impact of urbanization on bats. By identifying essential habitat features, such as vegetation cover and water availability, we can promote urban designs that are more conducive to bat populations. Furthermore, the study underscores the value of citizen science for ecological research, as it not only generates extensive data and empowers communities to participate in wildlife conservation but also enables researchers to conduct large-scale studies with limited resources, gathering data over broad geographic areas and fostering public interest in conservation efforts.

## BAT MIGRATION OVER THE GULF OF RIGA

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*Keywords: bat migration, bat behaviour, migration routes*

The Baltic Sea coast is a crucial migration route for many bat species. More specifically, a recognized migration route for bats, possibly originating from Finland, the Baltics, and Russia, extends along the eastern coast of the Gulf of Riga. While acoustic and physical evidence from Kabli and Pape ornithological stations provide data on the coastline route, evidence for migration over the Gulf of Riga is currently limited to anecdotal information. This study presents the first comprehensive data to assess bat activity patterns above the Gulf of Riga and the associated weather conditions.

We deployed 12 Batcorder devices from May 1, 2023, to October 10, 2023, positioned offshore in the Gulf of Riga. They were situated west and south of Kihnu island (Estonia), spanning distances of 13 to 32 km from the nearest mainland point or Kihnu. Bat recorders were affixed to buoys crossing potential flight paths from Estonia's west coast to the Gulf's west coast. Additionally, we conducted boat transects on 14 nights in spring and 28 nights in autumn. Concurrently, bat monitoring devices were deployed on Kihnu island and Kabli on the Gulf's east coast to facilitate comparisons of migration timing.

Altogether, during the study, we recorded 121 bat passes over the sea and identified five species: *Eptesicus nilssonii*, *Vespertilio murinus*, *Nyctalus noctula*, *Pipistrellus nathusii*, and *Myotis dasycneme*. The most abundant species was *Pipistrellus nathusii*, with 47 passes, followed by *Nyctalus noctula* and *Vespertilio murinus*, with 16 and 10 passes, respectively. Unfortunately, 33.9% (41) of the passes were only identified to higher taxonomical levels (primarily as *Nyctalus/Eptesicus/Vespertilio*).

Most bat passes were recorded during the autumn migration, totaling 103 (1.24 passes per 100 hours). In contrast, bat activity was significantly lower during the spring and summer, with 16 (0.92) and 2 (0.05) bat passes recorded, respectively. Migration predominantly occurred under calm weather conditions, favouring nights with wind speeds up to 5 m/s and no precipitation. Bats were also more likely to migrate with warmer temperatures.

We conclude that the Gulf of Riga is a seasonal bat migration route, with more bats likely migrating during fall. The migration is strongly weather-dependent, and bats prefer lower wind speeds and warmer temperatures over random nights. Areas far from the coast are seldom used during the summer period when bats are gathered in their summer colonies.

# GENETIC DIVERSITY AND PHYLOGEOGRAPHY OF THE ENDANGERED SIBERIAN FLYING SQUIRREL (*Pteromys volans*) OVER ITS RANGE

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*Keywords: genetic diversity, mtDNA, phylogeography, Siberian flying squirrel, SNP genotyping*

The Siberian flying squirrel (*Pteromys volans*) is an arboreal rodent with a distribution range that covers large parts of the Eurasian taiga forest zone. However, extensive forestry has resulted in widespread local population declines and extinctions in recent decades. In the European Union, vast declines have led the species to be listed in the Annexes of the EU Habitat Directive. As Finland has the largest population of Siberian flying squirrel in Europe, it has a particular responsibility for this species' conservation. Genetic diversity and sufficient gene flow are important for a species' ability to cope with changes in the environment in the long-term. Thus, understanding phylogeography and genetic diversity over the whole range helps to relate patterns also observed on a smaller scale. In this study we used genome-wide single nucleotide polymorphisms (SNPs) and mitochondrial DNA (mtDNA) to investigate population structure, connectivity, and genetic diversity in different sites throughout the Siberian flying squirrel distribution range. Our results showed that the species has overall low nucleotide diversity and heterozygosity. Locations in Finland, on the western edge of the distribution, had the lowest diversities in both genomic SNPs and mtDNA, while individuals in the Far East (Sikhote-Alin, Russia) show the highest diversity. These findings fit with a rapid range expansion from the Far East to the west. We also found a strong genetic differentiation between Sikhote-Alin and all other locations investigated. Several analyses clearly showed a separate genetic lineage, which might warrant a revision of the taxonomic classification of this species. Moreover, from conservation point of view, the inferred very low genetic diversities at the western edge of the distribution are especially worrisome as those populations are currently experiencing strong declines and major habitat changes, which can be especially detrimental when standing variation is low.

# POPULATION GENETICS OF THE SIBERIAN FLYING SQUIRREL IN FINLAND IS SHAPED BY GLACIAL RETREAT AND LAND UPLIFT DURING THE HOLOCENE

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**Keywords:** *Siberian flying squirrel*, mitochondrial genome, phylogeography, population genetics, Finland

The Siberian flying squirrel, *Pteromys volans*, is the only member of the flying squirrel tribe (Pteromyini) in Europe and its populations have been steadily declining. Finland holds the largest population in the EU, ranging across the southern half of the country. Despite the large distribution, it was recently classified as “vulnerable” in the Finnish Red List after a population decline of over 30% in a single decade. Currently, not much is known about the genetic structure of this population. Due to low genetic diversity in the West-Eurasian phylogroup of *P. volans*, commonly used markers for population genetic studies such as cyt b turned out to be uninformative. Thus, we have used the higher genetic resolution of whole mitochondrial genome (mitogenome) sequences to analyze the population structure of *P. volans* across its range in Finland.

We present new whole mitogenome sequences (16,511 bp) of 54 *P. volans* individuals, 52 from Finland and two from Estonia, resulting in 41 haplotypes. A median-joining network analysis (including sequences of Korean *P. volans*, representing the Far East phylogroup) uncovered three population clusters within Finland.

Star-shaped networks and population genetic analyses (Fu’s *F* and Tajima’s *D*) indicate recent expansions for two of those clusters, which geographically cover southwest or northeast Finland respectively. Individuals of the third cluster are geographically aggregated in central Finland with singular individuals found across all of Finland. Additionally, the Estonian and Korean sequences connect closest to the third cluster. This indicates that the third cluster could represent the ancestral population which first colonized Finland after the Last Glacial Maximum (LGM).

The results give a first indication of the phylogeographical history of *P. volans* at its westernmost distribution range in the early Holocene. We hypothesize initial colonization of *P. volans* after glacial retreat and forest succession in Finland (approx. 11,700–10,300 years ago). During the LGM the weight of the ice sheet depressed the Earth’s crust and large parts of Finland remained underwater after the ice sheet retreat. As the crust rebounded (land uplift), relative sea levels dropped, exposing new habitats (approx. 10,300–9,000 years ago). Thus, we propose rapid expansion events occurred as land emerged and suitable habitats connected, shaping the present-day genetic structure of *P. volans* in Finland.

# POTENTIAL ROLE OF GENETICALLY INFORMED EX-SITU CONSERVATION FOR THE SIBERIAN FLYING SQUIRREL (*Pteromys volans*)

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*Keywords: ex situ conservation, breeding program, reintroduction, genetic monitoring, one plan approach*

The Siberian flying squirrel (*Pteromys volans*) is not only the sole representative of its tribe Pteromyini in Europe but also an indicator species for old-growth forests. The overall declining populations led to local extinctions during the last centuries like most recently in Latvia. Finland holds the largest population, while in the Baltic states, this enigmatic rodent can only be found in a relict population in Estonia. Despite important ongoing conservation efforts, the population decline is continuing and *P. volans* is threatened to disappear from European forests entirely. Successful species conservation often must combine several strategies to be successful. The desire to build up an ex-situ population with the potential for reintroductions was expressed by different sides and comparable to efforts in other species like Eurasian lynx, wisent, common hamster or European ground squirrel.

Every modern ex-situ program should be accompanied by a genetic evaluation. Our research with whole-genome data on population structure and genetic diversity of the Siberian flying squirrel paves the way for high-resolution molecular tools to enable evidence-based decision-making by conservationists. Genetic screening of the current captive population should significantly contribute to the selection of founders and follow-up management for ex-situ breeding programs. For the assessment and to consider all the needs of such a program, we teamed up with zoos, private holders, and umbrella organisations like the European Association of Zoos and Aquaria (EAZA). Genetic screening of the captive population is executed together with our ongoing genetic monitoring of wild populations in line with the One Plan Approach (OPA), which comprises the combined management of ex-situ and in-situ species conservation. At the same time, we try to document valuable knowledge about this species' husbandry to optimise potential breeding programs. With our partners, we are aiming to find new breeders to increase the limited holding capacity for this species.

When it eventually comes to reintroduction efforts, both ex-situ and in-situ populations could be managed together. This maximises the potential genetic diversity that is important for the long-term existence of wild populations. This talk invites the audience and the conservation as well as research community to engage with us to be able to develop a concept with the prospect of success for Europe's only native flying squirrel.

# GLIDING TOWARDS AN UNCERTAIN FUTURE: OPPORTUNITIES OF GENOMIC TOOLS FOR THE CONSERVATION OF SIBERIAN FLYING SQUIRRELS (*Pteromys volans*)

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*Keywords: Siberian flying squirrel, conservation genomics, population connectivity, conservation management*

Environmental and human-mediated changes are threatening biodiversity worldwide. Several species with key functions in their ecosystems are strongly declining or even going extinct. One such species is the Siberian flying squirrel (*Pteromys volans*). Within the European Union it is only found in Finland and a small population in Estonia, and its numbers are declining rapidly. Several conservation and monitoring projects are under way to protect this umbrella species, relying mostly on non-genetic methods. However, genetic methods can provide additional information, which are difficult or impossible to obtain with traditional approaches.

Here, we outline how genomic inferences and techniques can support conservation and traditional monitoring efforts. In order to develop efficient monitoring tools and to obtain high-resolution data on genetic diversity, inbreeding and connectivity within and between Siberian flying squirrel populations, we assembled a chromosome-level reference genome for this species and generated whole-genome data for over 50 individuals from Finland and Estonia. These data are being used to develop a reduced genetic marker set for large-scale monitoring using non-invasively sampled droppings. This can provide invaluable insights on the impact of landscape features, such as roads or clearcuts for forestry or powerlines on the connectivity of populations. Furthermore, we developed non-invasive methods for inexpensive molecular sex determination and molecular diet analyses based on droppings obtained from traditional monitoring efforts.

Recently, the gap between science, society and policy has been highlighted as a major problem for developing and running conservation projects and long-term global change mitigation strategies. To bridge this gap our project is run together with several national and international stakeholders and collaborators, and the public.

## GENETIC CHARACTERISTICS OF EUROPEAN BISON (*Bison bonasus*)

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**Keywords:** European bison, genetic variability, microsatellites, mtDNA, SNP

European bison (*Bison bonasus*), the largest terrestrial mammal in Europe, remains critically dependent on effective conservation management due to its narrow genetic base and population fragmentation. This study investigates the genetic diversity of European bison populations in Lithuania, comparing them with populations from other European countries to provide insights into their genetic diversity.

We analysed genetic variability using three molecular markers: microsatellites, mitochondrial DNA (mtDNA), and single nucleotide polymorphisms (SNPs). Microsatellite markers were employed to assess nuclear genetic diversity, population structure, and inbreeding levels, while mtDNA provided insights into maternal lineage variability. Additionally, SNP genotyping enabled high-resolution detection of genetic lines and genetic differences between populations.

Our findings indicate that the Lithuanian population has at least two different haplotypes, retains low genetic diversity, however, the genetic structure is different compared to other European populations. The comparison of SNP-based population structure reveals genetic line related clusters, reflecting geographical and historical factors influencing genetic makeup. These results highlight the need for genetic management, such as controlled translocations, to enhance connectivity and minimize inbreeding risks.

This study contributes to understanding the genetic characteristics of European bison, emphasizing the importance of integrating genetic data into conservation strategies to ensure the species' long-term viability across its range.

## IDENTIFYING WOLF-DOG HYBRIDS IN ESTONIAN WOLF POPULATION WITH A SNP PANEL

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**Keywords:** wolf (*Canis lupus*), dog (*Canis familiaris*), hybridization, introgression, Estonia, SNP genotyping

Hybridization is the process of mating between two species or subspecies that can result in viable offspring. It can pose a threat to the genetic integrity of a species if followed by introgression. The identification of hybridization and introgression in wolf populations is important because these processes can reduce the fitness, survival and long-term persistence of the population. In this Master's thesis, a new generation marker system, a panel of 96 SNP markers was used for the first time in Estonia to identify wolf-dog hybrids. The programs PCA, ADMIXTURE, HYBRIDLAB, NewHybrid and ML-Relate were used to identify the hybrids and determine their relatedness. While in previous work backcrosses up to generation 1 have been determined, in the present work it was possible to identify hybrids up to generation 4 and to find relatedness between them, indicating that at least two hybridization events occurred. A total of 9 hybrids were identified, one of which was a second-generation hybrid, two of which were third-generation and six of which were fourth-generation hybrids, all were backcrossed into the wolf population. The rate of introgression in our study was ~ 1.64%.

# POPULATION GENETICS OF THE GREY WOLF (*Canis lupus*) AND THE INFLUENCING FACTORS IN ESTONIA

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*Keywords: Canis lupus, population genetics, gene flow, cross-border genetic exchange*

The grey wolf (*Canis lupus*) is a large carnivore and apex predator, playing a critical ecological role in its ecosystem. Previous studies have examined various aspects of grey wolf populations, including genetic diversity. In the Baltic region, the heterozygosity of local wolf populations remains relatively high compared to other regions in Europe, a finding supported by several prior studies.

The primary aim of this research is to assess the current population dynamics of the Estonian wolf population and to evaluate cross-border gene flow, particularly from Russia. This is of critical importance because, while a large population size may appear to ensure genetic diversity, it may not be sufficient to maintain it if gene flow is restricted. Previous studies have suggested that the heterozygosity of the Estonian wolf population is high, and this forms one of the hypotheses of our research. A second hypothesis posits that genetic material from Russia is currently flowing into the Estonian population through wolf movement. This new study is needed due to recent changes in landscape structures, such as the construction of border fences along the Estonia-Russia border, which could restrict wolf movement. Additionally, the significant hunting pressure in Estonia, one of the highest in Europe, may further affect the genetic health of the population.

In this study, we used the Thermo Scientific Canine Genotypes Panel 1.1 to genotype individuals from tissue samples collected through legal hunting and fecal samples collected between 2015 and 2024, analyzing 16 microsatellite loci.

This research is crucial for the effective conservation and management of wolf populations, as it provides essential data on the genetic condition of the Estonian wolf population. The findings will serve as a baseline for future studies, particularly given the potential expansion of border fences along the Estonia-Russia border and the ongoing hunting pressures, which are unlikely to decrease in the near future.

## WHAT CAN WE LEARN FROM GENETIC MATERIAL OBTAINED FROM HUNTED GREY WOLVES IN LATVIA?

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*Keywords: wolf, hunting, genetic monitoring, social and kinship structure*

In Latvia, the grey wolf (*Canis lupus*) is a specially protected species of limited use. Since 2004, an annual quota has been set, and a wolf hunting season has been established. The annual quota and number of hunted individuals have grown from 130–200 to 280–300 wolves in a hunting season. Although the sampling of only hunted wolves has drawbacks and limitations, the large amount of available data makes it possible to gain valuable information on the genetic and kinship structures and social dynamics of the Latvian wolf population.

Muscle tissue samples from legally hunted or otherwise killed (traffic accidents, scabies) wolves have been collected between July 15, 2009 and March 31, 2024. A total of 2,063 individuals were successfully genotyped, accounting for 53.1% of all wolves harvested during this period. The genetic parameters of the population were calculated using the standard parameters in the GenAEx 6.5 program. Kinship analyses were performed using the COLONY 2.0.5 and CERVUS programs.

Analyses of the data obtained from hunted wolves have provided information on the average expected heterozygosity ( $H_e$ ) ( $0.732 \pm 0.019$ ), the observed heterozygosity ( $H_o$ ) ( $0.707 \pm 0.020$ ), the inbreeding coefficient ( $F$ ) ( $0.034 \pm 0.008$ ) and allelic diversity (ranging from 5.81 to 6.39) of the entire population. Comparing data between the eastern and western parts of the country, no significant differences were found between several genetic parameters, and these parts of the population were not genetically differentiated, thus confirming that the Latvian wolf population is not fragmented. Kinship analyses have identified a total of 277 kin groups and allowed the establishment of their longevity and territorial distribution. It was determined that two groups had existed for at least 12 years, while most kin groups (66.8%) were detected for only up to four years. Insight into the history of some packs and the dispersal and migration of kindred individuals was gained. Breeder loss, disruption of packs, territorial displacement, and early dispersal of juvenile wolves were determined in some of the groups.

## DID LITHUANIA EXPERIENCE THE ANTHROPAUSE IN TERMS OF ANIMAL ROADKILL DYNAMICS?

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*Keywords: COVID-19 lockdown, roadkill dynamics, urban wildlife collisions, anthropause effects*

Studies of roadkill in Lithuania during the COVID-19 pandemic confirmed the effects of reduced human mobility and increased wildlife activity. The lockdown measures, characterized by strict travel restrictions, resulted in significant reductions in roadkill on major highways, with reductions of up to 75–90% during the peak lockdown periods of April–May 2020 and November–December 2020. These reductions reflected the immediate impact of fewer vehicles on the road. Conversely, urban areas experienced a sharp increase in roadkill, with incidents involving roe deer nearly doubling from 700 in 2019 to 1,281–1,325 in 2020–2021. This trend resulted in an overall increase in urban roadkills of 55% in 2020 and 29% in 2021, compared to expectations based on historical trends. These changes highlight the contrasting effects of reduced traffic on major roads and increased wildlife presence in urban environments during the pandemic.

The dominance of roe deer in these events was striking, with the species accounting for 68.6% of urban roadkill. In addition, domestic animals, including dogs and cats, accounted for 12.2% of urban roadkill, reflecting a unique dynamic in urban environments. Such increase in urban roadkill is likely due to changes in animal movement patterns as wildlife ventured into quieter urban spaces during closures, coupled with the continued operation of essential transportation in urban areas. This phenomenon, called “anthropause”, temporarily altered human-wildlife interactions. Wildlife activity increased during COVID, but the long-term benefits for wildlife were mixed. While reduced traffic initially provided a reprieve for animals on highways, the increased presence of wildlife in urban areas, coupled with resumed mobility after restrictions, led to increased collision risks. By 2022, roadkill numbers in both urban and rural areas began to return to pre-pandemic trends, underscoring the transient nature of these shifts.

Our findings highlight the need for proactive and adaptive protection strategies to mitigate future risks. Improved urban road planning, the creation of wildlife corridors, and increased driver awareness campaigns are essential, especially during periods of changing human activity.

## ATTITUDES TOWARDS LARGE CARNIVORES IN BALTICS: SWITCH FROM POST-SOVIET RURAL INTO MODERN URBAN SOCIETY

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*Keywords: attitude towards large carnivores, stakeholders, human-wildlife relations, wolf, lynx, bear, Europe, Baltic countries*

The ongoing increase in populations of large carnivores (grey wolf, brown bear, European lynx) and their co-occurrence with humans in European human-modified and agricultural landscapes has increased controversies and conflicts. The range of human attitudes toward these charismatic species is broad, varying from admiration to hate and fear. Recent research has emphasised the need for sustainable carnivore management programs that integrate not only biological or ecological but also social, cultural and political components. Given that any policy is ultimately shaped by societal values and attitudes, successful coexistence with large carnivores thus requires public policies that account for people’s attitudes. The main goals of this study were to identify (1) the existing attitudes of society (including both the general public, but also specific segments, such as hunters, farmers, bee-keepers) towards large carnivores at the present moment, and (2) compare it to the previous study period (2003–2006). We carried out our study during the period of 2022–2023 in Estonia, Latvia and Lithuania by distributing an electronic version of the questionnaire through both schools and other sources (such as local government general contacts and their newspapers and local newspapers, Facebook groups, etc). The questionnaire design was based on a previous study done in the mid-2000s. It contained five different sections (I – Large carnivores and society; II – Wildlife and society – Wildlife value orientations and attitude; III – Special questionnaire for hunters; IV – Special questionnaire for farmers and/or beekeepers; V – General Information), altogether 49 questions on attitudes, beliefs and knowledge toward large carnivores, specialised questions for hunters, farmers and/or beekeepers, as well as on the demographic information of respondents. In the presentation, the main findings on attitudes towards large carnivores and changes in those attitudes compared to the previous study period, both on species and country level in three Baltic countries are presented.

# WOLF PREDATION ON LIVESTOCK MANAGEMENT INSIGHTS IN LITHUANIA

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*Keywords: wolves, feeding strategies, domestic animals, individual behavior, Lithuania*

This study analyzes the behavior of wolves (*Canis lupus*) in Lithuania, where recent years have seen an increase in the populations of wild prey, such as red deer (*Cervus elaphus*) and roe deer (*Capreolus capreolus*). As a result of these changes, hunting quotas for these species have been increased, providing wolves with sufficient food in the wild. Despite this, some wolves continue to attack domestic animals, creating conflicts with farmers. The aim of this study was to investigate the reasons behind the emergence of “problematic” wolves and to identify their behavioral characteristics.

The research methods included site analysis, examination of affected livestock, tracking wolf activity, and reviewing video footage obtained from cameras set up at livestock attack sites. The findings emphasize the need for individualized preventive measures to reduce conflicts between humans and wolves. The study advocates for the implementation of more effective protection systems and enhanced cooperation between farmers and conservation organizations. The collected data can be used to inform policies aimed at harmonizing human-wolf co-existence while maintaining ecological balance.

During the 2022 study, it was found that even with an adequate supply of wild prey, certain wolves prefer domestic livestock as an alternative food source. In 2024, the research was extended to analyze five wolf attacks on domestic animals. The results revealed that, in all cases, the attacks were carried out by a single wolf, not by a wolf pack. This indicates that the issue is linked to individual adaptive mechanisms rather than group behavior.

## FEAST OR FAMINE: NAVIGATING HERBIVORE IMPACTS ON REGENERATION IN HEMI-BOREAL FORESTS

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*Keywords: ungulates, biodiversity conservation, forest gap, regeneration, browsing*

Increased commercial forest harvesting and young forests as well as predator control can stimulate increased herbivore populations. The cascades from these management decisions may lead to multiple effects on composition, structure and on critical ecological forest processes, such as succession and tree recruitment.

The aim of this study was to analyze browsing pressure in mixed coniferous forests using natural gaps in commercial and nature reserves (protected areas). We hypothesize that animals stay in protected forests more than in commercial ones, which is correlated to increased browsing damage and reduced recruitment of trees.

We compared the differences in herbivore effects on both commercial forests and forest reserves by measuring browsing damage on young saplings (1–4 m in height) in naturally formed forest gaps, the amount of tree layer food, and immediate herbivore density near gaps and in forests by pellet counts.

The results indicate that ungulates, both in commercial forests and reserves, are damaging almost all regeneration saplings, with slightly more damage observed in commercial forests due to the higher concentration of saplings after clearcuts. In terms of ungulate density, reserves had more animals, compared to commercial forests.

**Conclusion:** The study concludes that ungulate damage to forest regeneration is a significant issue in both reserves and commercial forests. However, the damage is slightly more pronounced in commercial forests due to the higher overall concentration of saplings. Therefore, forest management actions are needed to help recruitment of young saplings into the forest population. Further research is needed to explore mitigation strategies and to compare these findings with other regions.

## MONITORING OF DEER DAMAGE TO YOUNG PINE, SPRUCE AND ASPEN STANDS, 2018– 2023. DATA OVERVIEW

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*Keywords: National forest inventory, pine stands, spruce stands, aspen stands, ungulates*

The monitoring of deer damages to young pine, spruce and aspen stands in Latvia is has been implemented since 2014, and is a part of biotic risk monitoring in the National forest inventory. The aim is to obtain information about damages caused to forest stands by deer and their population density and to predict threats to forestry. 200 stands of each tree species (pine, spruce, aspen) are surveyed each year. The monitored pine and aspen forest stands are up to 20 years old and spruce stands – up to 40 years old. If the characteristics of an initially selected stand change during the study period, e.g. due to growth, preventing deer from reaching the edible branches or bark, it is substituted with the closest appropriate stand. Field surveys are performed after the complete disappearance of snow cover which usually occurs in April.

All data are collected in circular sample plots (area 100 m<sup>2</sup>), which are distributed regularly within each selected pine stand to represent 5% of each selected stand area (a minimum of 4 sample plots per stand). The following data are collected: (i) the condition of the pine, spruce and aspen trees based on the presence of fresh damages (undamaged, lightly damaged, heavily damaged, destroyed or dead because of previous damages); (ii) the average height of the dominant trees; (iii) the number of moose, red deer and roe deer pellet groups (in order to obtain a site-specific, comparable relative deer density index). The species, number and average height of the admixed tree species and undergrowth plants are obtained, information about individual tree protection measures used in the stand, as well as the presence of fresh pre-commercial thinning, is recorded.

The number of young stands in which the average proportion of severely damaged and destroyed trees exceeds 1% has been fluctuating, for pine stands it was on average 55% of all surveyed stands. In young stands up to 2 m in height and also from 3–4 m in height, the average proportion of damaged stands annually is slightly over 60%, while in young stands with a higher height of the dominant species, the proportion of damaged stands is around 37% of all pine stands surveyed in the respective height group. The number of damaged spruce stands is small – in the period under review it fluctuates from 12.82% to 25.6% of all surveyed spruce stands. The number of damaged aspen stands in this time period also fluctuates from year to year, but on average it is 43% of all stands. Although the proportion of fresh damages in repeatedly surveyed young stands decreases every year, the impact of the damages is permanent in the long term.

## CAN THE PRESENCE AND STRUCTURE OF UNDERGROWTH AFFECT THE RISK OF DEER DAMAGE TO YOUNG PINES?

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*Keywords: pine stands, undergrowth, pre-commercial thinning, deer damages*

Despite the fact that deer populations in Latvia have been decreasing slightly in recent years, they remain high and pose a significant risk of damage to young stands of economically important tree species. Diverse admixed tree species and undergrowth plants in stands can provide food for all ungulate species, especially in winter. As their diet in the cold season largely overlaps, the availability of preferred food and animal density are factors that significantly determine the risk of damage to target species in young stands. Summer browsing of woody plants can indicate the potential risk of damage to the dominant species in the following winter: the more intensively these sites are grazed during the vegetation period, the higher the risk of damage in the following winter.

We used data collected in the year 2023 within the framework of the National Forest Inventory in Latvia, specifically – ‘Deer damage to young pine, spruce and aspen stands’. All data were collected from 206 pine stands up to 20 years of age by setting up a total of 1,454 circular sample plots (area 100 m<sup>2</sup>) distributed regularly within each selected pine stand. The following data were collected: (i) the condition of the trees based on the presence of fresh damages; (ii) the average height of the dominant trees; (iii) the species, number and average height of the admixture and undergrowth; and finally; (iv) the number of moose, red deer and roe deer pellet groups (in order to obtain a site-specific, comparable relative deer density index). The presence of fresh pre-commercial thinning was recorded. The information about pine stand forest types were gathered from Forest State register database. In addition to previous information, the proportion of ungulate summer and winter browsing of undergrowth in 60 pine stands was assessed in late summer 2023 and spring 2024.

Results show that in pine stands with dominant height below 3 m, the probability of damage decreases with an increase in the density of undergrowth for ‘dry’ and ‘wet’ forest types. The proportion of pine damage in freshly managed young stands was higher than in unmanaged young stands,  $9 \pm 1\%$  and  $5 \pm 0.4\%$ , respectively. Moose and roe deer density index were higher in un-thinned stands whereas for red deer – in thinned stands. Analyzing the relationship between the intensity of summer browsing in the previous (2023) vegetation period and the proportion of pine tree damage in the winter of 2024, a significant positive relationship was observed – in young pine stands with a higher proportion of undergrowth damage in the previous summer, the proportion of pine trees damaged in the winter is also higher. Our results indicate the importance of the presence of undergrowth in reducing deer damage to young pine stands.

## **WANTED DEAD OR ALIVE – WILD BOAR *Sus scrofa* CONTROL CHALLENGES CONCERNING AFRICAN SWINE FEVER OUTBREAK IN LATVIA**

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*Keywords: wild boar, African swine fever, population control*

Over the last half-century, the wild boar has been a game species of particular importance to the local economy and rural social life in Latvia, i.e. one of the so-called “big five” for recreational hunters at a national scale. The increase in their abundance was deliberately promoted by avoiding hunting of mature female boars, supplementary feeding and control of predators, namely wolves. Only briefly hindered by local and temporal outbreaks of controllable classical swine fever in the 1990s, the wild boar population grew to its maximum, reaching 74,000 estimated and 37,000 hunted individuals by 2013.

While not speculating that overpopulation was a cause for the African swine fever (ASF) outbreak in the wild boar population in Latvia, the disease was first detected in June 2014 during a routine surveillance procedure of animal carcasses found dead in the natural environment. In addition to the immediately strictly implemented biosecurity measures in domestic pig farming and pork industry, carcass search and removal in the wild, relevant measures were taken to reduce wild boar numbers and population density. A drastic reduction of population size was soon reported locally and also a three-fold decline at the country scale, however, compared to historical data, the numbers still are at least twice as high as before 1970 when the wild boar population increase started.

Currently, further depopulation is crucial because ASF has endemically established in the wild boar population and natural environment while mass mortality repeatedly occurs in places where populations have recovered. The methodology of wild boar census and a system for controlling numbers and population density is proposed by a study. By establishment of cooperation with the European Observatory of Wildlife, which supports and coordinates the monitoring of wildlife populations in Europe according to a common protocol for the use of trail cameras, a pilot census in the central part of the country was performed. In parallel, an analysis and comparison of data on wild boar sex and age from hunting bags at feeding sites and during driven hunts was performed to demonstrate the interaction between population structure and dynamics. First results and considerations for management feasibility are summarized. Hence, finding a way to introduce our findings to the hunting community and to motivate them for management actions is another challenge of a system implementation to prevent the spread of ASF.

## ROLE OF MAMMALS IN TRANSMISSION OF VECTOR-BORNE PARASITES AND PATHOGENS

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*Keywords: mammals, vector-borne pathogens, parasites*

Wild mammals in Europe play a significant role as vectors and reservoirs of vector-borne pathogens (VBP).

The occurrence of vector-borne diseases is increasing globally, which represents a considerable threat to public health and veterinary medicine. Ticks, mosquitoes and VBP have co-evolved with wildlife. Wild ungulates, hedgehogs, small- and medium-sized carnivores, and squirrels are important feeding hosts for ticks and serve as reservoirs for tick-borne pathogens.

The distribution areas of wildlife, ticks, mosquitoes and pathogens they carry are undergoing significant changes due to anthropogenic impacts such as urbanization, habitat fragmentation, changing land-use patterns, animal trade and travel. Different mammals play different roles in the transmission of VBP: some are important components in the developmental cycle, while others may act as dilution hosts (cervids for *Borrelia burgdorferi* (*sensu lato*) and tick-borne encephalitis virus). The objective of this review is to provide a concise overview of the most important and emerging VBPs from a One Health perspective and to identify knowledge gaps regarding the involvement of wild mammals in their endemic cycles.

## BROWN BEAR (*Ursus arctos*) ENDOPARASITES AND THEIR SEASONAL DYNAMICS IN ESTONIA

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*Keywords: brown bear, parasites, parasitofauna, Baylisascaris transfuga, flotation*

The brown bear (*Ursus arctos*) is an omnivore and the largest member of the order Carnivora in mainland Europe, which is why it needs a lot of food, both plants and animals. But eating large volumes of diverse food can lead to contacts with various parasites. As an animal with good movement and a relatively long lifespan (20–30 years), the bear can both be a long-term reservoir and an extensive spreader of parasites. Despite the significant role of parasites, knowledge of their diversity in the brown bear is still scarce and current research on wildlife parasites in Estonia has covered several other mammal species, such as the red fox, raccoon dog, golden jackal, grey wolf and Eurasian lynx, while data for the brown bear has been absent. In Europe, researchers in multiple countries have researched the parasitofauna of the brown bear, but no research has been published from Eastern or Northern Europe. The aim of this work was to fill the knowledge gap regarding parasites spread by brown bears in Eastern and Northern Europe and assess the role of bears in spreading parasites that can also be dangerous to humans.

For this, we collected bear scats mainly from the brown bear core area of Alutaguse in North-Eastern Estonia, confirmed the host species via DNA sequencing and examined the parasite eggs present in the scats via flotation analysis. Out of 148 samples, 75% contained at least one parasite egg, which is more than in most previous research done in Europe (0–79%). Protozoan eggs (*Eimeria*, *Giardia*, *Coccidia*), were found in 16% of samples, 7% contained cestode (*Taenia*, *Diphyllobothrium*) eggs, 60% contained nematode (*Strongylidae*, *Ascarididae*, *Capillaria*, *Baylisascaris transfuga*) eggs and 4% contained trematode eggs. The yearly prevalence for the more researched bear-specific nematode *B. transfuga* was 51%. Compared to other European research, no eggs of *Ancylostoma* and *Uncinaria* were detected, and one *Acanthocephala* egg was detected. The general seasonal dynamic for the parasite eggs followed the findings of previous research, wherein the smallest number of samples contain parasite eggs in spring and the most in the fall. *B. transfuga* also followed this seasonal dynamic. The opposite dynamic appeared with protozoa, which were more frequently present in samples in spring and in the least number of samples in the fall, which may point towards an antagonistic interaction between protozoa and nematodes.

## **TOXOPLASMA GONDII PARASITES IN THE EURASIAN BEAVER (*Castor fiber*) IN LITHUANIA**

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*Keywords: Toxoplasma gondii, beaver, PCR*

A total of 79 brain samples from Eurasian beavers (*Castor fiber*) were analyzed to detect the presence of *Toxoplasma gondii*, a significant zoonotic protozoan parasite. Using nested polymerase chain reaction (PCR) targeting the ITS1 region, followed by sequencing, 8 samples (10.13%) tested positive for *Toxoplasma gondii*. Positive samples were further analyzed using Sanger sequencing and quantitative PCR (qPCR). Age structure analysis revealed that infections were observed in beavers with an average age of 5 years, with males being predominant among the infected individuals (5 out of 8 samples).

The beavers were collected from the central and eastern regions of Lithuania through legal hunting methods. This study represents one of the first investigations of *T. gondii* in Lithuanian rodents and underscores the importance of understanding the genetic diversity, genotypes, and sub-genotypes of *T. gondii* circulating in the region. These findings provide a valuable baseline for further research into the prevalence and genetic characterization of this zoonotic pathogen in rodent populations in Lithuania.

## PREVALENCE OF VECTOR-BORNE PATHOGENS IN EUROPEAN BISON (*Bison bonasus*)

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**Keywords:** European bison, tick-borne pathogens, Lithuania, Poland

Wild ungulates in Europe play a significant role as vectors and reservoirs for tick-borne pathogens, including *Anaplasma* spp., *Bartonella* spp., *Babesia* spp., and *Rickettsia* spp. The European bison (*Bison bonasus*), Central Europe's largest and rarest wild ungulate, exists in both captive and free-ranging populations. Over the past two decades, the size and distribution of free-ranging European bison populations have grown, resulting in increased interactions with both wild and domestic animals and potentially heightening the risk of pathogen transmission. However, due to the European bison's protected status, research on tick-borne pathogens in this species remains challenging and relatively limited. The aim of this study is to investigate tick-borne pathogens in bison spleen samples and ticks collected from bison in Lithuania and Poland. A total of 85 spleen samples from European bison and 560 ticks (comprising 408 *Ixodes ricinus* and 152 *Dermacentor reticulatus*) were analyzed. Tissue samples and ticks collected from the bison were screened for *Anaplasma*, *Babesia*, and *Bartonella* species using RT-PCR targeting various gene loci. RT-PCR results showed infection rates of 40% for *Anaplasma*, 7% for *Babesia*, and 7.9% for *Borrelia* spp. All positive samples were further tested using nested PCRs with a set of genus-specific primers. Sequence analysis of the partial 18S rRNA gene identified *Babesia divergens* and *Babesia capreoli* in European bison, while ticks carried *Babesia divergens*, *Babesia microti*, and *Babesia venatorum*. Additionally, the analysis revealed two unique 16S rRNA variants, six *msp4* variants, and nine *groEL* variants of *Anaplasma phagocytophilum* in European bison and their associated ticks. Phylogenetic analysis of 16S–23S rRNA ITS, *gltA*, and *rpoB* partial gene fragments confirmed infections of European bison with *Bartonella bovis* and *Babesia schoenbuchensis*. Given the endangered status of European bison, further research is essential to understand the potential health impacts of these tick-borne infections.

## THE EUROPEAN BADGER AS A POTENTIAL HOST FOR VECTOR-BORNE PATHOGENS IN LITHUANIA

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*Keywords: Meles meles, Anaplasma phagocytophilum, Babesia spp.*

The aim of the present study was to investigate the occurrence of vector-borne pathogens in European badgers (*Meles meles*) from Lithuania. DNA was extracted from spleen samples by using a Genomic DNA Purification Kit, according to the manufacturer's instructions. Badgers were screened via PCR using specific primers for *Anaplasma* sp., *Bartonella* spp., *Borrelia* spp., *Rickettsia* spp. and *Babesia* spp. Positive PCR products were selected for DNA sequencing. The results of this study revealed that 28.5% of the badgers were infected with *Anaplasma* sp. and 71.4% were infected with *Babesia* spp. However, *Bartonella* spp., *Rickettsia* spp. and *Borrelia* spp. were not detected in any of the tested samples. Phylogenetic analysis showed that the sequences obtained from badgers clustered closely with *Anaplasma phagocytophilum* and known *Babesia* species recently identified in badgers. This is the first report indicating such a high prevalence of these vector-borne pathogens in European badgers from Lithuania. However, further research is required to investigate other potential pathogens that may be harbored by European badgers, as their role in the epidemiology of vector-borne diseases may extend beyond the pathogens identified in this study.

## INTERACTIONS OF HELMINTH PARASITES AND CARNIVORE MAMMALS IN A CHANGING ENVIRONMENT: RED FOX AS A CASE STUDY

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*Keywords: red fox, helminths, zoonotic parasites, landscape*

Red fox (*Vulpes vulpes* L.) is a widely distributed, common, and abundant canid. This predator is found in various habitats and is well adapted to environmental changes caused by humans. At the same time red fox serves as reservoir host of various zoonotic parasites. The aim of the study was to determine the diversity of helminth parasites (trematodes, cestodes, and nematodes) and to estimate the prevalence of infection in different landscapes. Red fox faeces were collected in three different landscapes (forest, fragmented and agricultural) from 2023 to 2024 in Lithuania. Two techniques for detection of parasitic helminth eggs were applied: floatation for trematodes and sedimentation for cestodes and nematodes. Two species of trematodes and six nematode species were identified. Meanwhile, cestodes were identified to genus and family levels (as more accurate identification was not possible using light microscopy). In all landscapes, helminth eggs were detected in more than 90% of fox faecal samples. The highest prevalence was estimated for the trematode *Alaria alata* (Goeze, 1792), followed by the nematodes *Eucoleus aerophilus* (Creplin, 1839), *Uncinaria stenocephala* (Railliet, 1884), and *Toxascaris leonina* (Linstow, 1902) Leiper, 1907.

## **PATHOLOGY OF TEETH AND MANDIBLES IN VOLES OF THE SUBFAMILY ARVICOLINAE**

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*Keywords: voles, Arvicolinae, dental and mandible pathology*

The aim of this study is to conduct a meta-analysis of the prevalence of dental and mandibular pathology in voles of the subfamily Arvicolinae. Shape and structural changes of teeth and lower mandible are a widespread phenomenon that has been found both in nature and in laboratory colonies. In the Palearctic, pathological heterochrony (excessive tooth growth, curvature, fractures, malocclusion) and mandible pathology (overgrowth of bone tissue, formation of cavities, etc.) were found in *Microtus arvalis*, *Agricola agrestis* (Bashenina, 1996; Jentzsch et al., 2020), *M. hartingi* (Zorenko, 2021), *Alexandromys montebelli* (Sugita et al., 1995) and *Lagurus lagurus* (Imbschweiler et al., 2011). In the Nearctic, similar dental pathologies have been reported in *Microtus californicus scirpensis* (Imai et al., 2018), *My pinetorum* (Harvey et al., 2009) and *Myochrogaster* (Jheon et al., 2015).

Various hypotheses are considered to explain the occurrence of bone pathologies in laboratory vole colonies. These include a soft diet (absence of solid timber suitable for abrasion of teeth), osteomyelitis as a result of infection, the development of benign tumors (hamartomas) as well as genetic background. We hypothesize that disturbances in the development of the lower jaw and the growth of the upper incisors could be the result of environmental stress. The possibility of genetic inheritance of this trait is considered, as well as the possibility of this side effect being caused by the combination of genetic and environmental factors (Gill & Bolles, 1982).

With stress and the bottleneck effect, associated with the extreme environmental changes, epigenetically determined disturbances in the formation of cranial bones and incisors heterochrony could occur during ontogenesis. Many traits may possess a hidden quantitative nature of variation, manifesting in individuals whose trait anlage exceeds a certain threshold value (Grüneberg, 1952). These disturbances have ancient origins, occurring in both Nearctic and Palearctic species whose ancestors diverged at the molecular level around 3.8–4 Ma ago (Abramson et al., 2021). Similar environmental stressors acting during a sensitive period of ontogenesis in voles can apparently cause the same developmental anomalies in different taxa.

## **ANTHROPOGENIC IMPACT ON GENETIC DIVERSITY OF RED DEER (*Cervus elaphus*)**

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*Keywords: red deer, genetic diversity, anthropogenic impact*

Red deer is a native species to Central Europe, but the areas where it lives sympatrically with the introduced sika deer have increased in the last three decades. Hybridization between the two species and changes in population genetic structure are the most important problems. Red deer stags may be attacked by sika males, which are extremely aggressive in the rutting season and may mate with red deer hinds, with the inherent risk of hybridization and introgression between the two species. The genetic diversity of the red and sika deer species was studied using tissue samples from legally hunted animals in Lithuania. Genotyping was based on microsatellite loci (STR) of nuclear DNA. Molecular genetic data combined with evaluation in statistical programs could lead to a complex view of populations and differences among them. The results of hybridization between the two species are documented in different EU countries. The anthropogenic impact of sika deer on red deer populations is discussed.

## **GENETIC STRUCTURE OF THE LITHUANIAN WILD BOAR (*Sus scrofa*) POPULATION AFTER THE OUTBREAK OF AFRICAN SWINE FEVER**

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*Keywords: wild boar, genetic structure, African swine fever, Lithuania, microsatellite*

This study aimed to assess how African Swine Fever outbreaks influenced the genetic diversity and structure of the Lithuanian wild boar (*Sus scrofa*) population. A total of 491 wild boar samples collected across Lithuania between 2014 and 2019 were analyzed. Genomic DNA was extracted, and samples were genotyped using 16 short tandem repeat (STR) loci. Results revealed allele richness ranging from 15 to 51 and significant deviations from Hardy-Weinberg equilibrium across all loci. Fixation indices showed a marked reduction in heterozygosity within and among subpopulations. Principal coordinates analysis (PCoA) and STRUCTURE clustering identified genetic differences between the ASF-non-affected western region (restricted zone I) and ASF-affected regions (restricted zones II and III). These findings suggest that environmental factors and disease dynamics have shaped regional gene flow and influenced the genetic structure of wild boar populations in areas affected by ASF.

## VECTOR-BORNE DISEASES OF COMPANION ANIMALS IN URBAN AREAS OF LITHUANIA

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**Keywords:** vector-borne diseases, *Babesia canis*, *Anaplasma phagocytophilum*, Lithuanian hound dogs, urban canine health

Vector-borne diseases represent a serious risk to both animal and human health in the Baltic region, including Lithuania, owing to the temperate climate, extensive forests, and high tick densities, particularly of *Ixodes ricinus* and *Dermacentor reticulatus*. These ticks thrive in urban areas, forests, and recreational green spaces, which are frequently visited by dogs and their owners, increasing the risk of exposure to tick-borne pathogens. According to the European Pet Food Industry Federation (FEDIAF), 75.8% of Lithuanian households own at least one dog. Data from FEDIAF and various Lithuanian institutions indicate that there could be a total of 550,000–580,000 dogs in Lithuania, a significant portion of which reside in Kaunas, the country's second-largest city. This study investigated the prevalence of tick-borne pathogens, specifically *Babesia canis* and *Anaplasma phagocytophilum*, in dogs visiting a veterinary clinic in Kaunas. Blood samples were collected from 203 dogs, including pet and hunting breeds (Lithuanian Hounds), during 2023–2024. Pathogen detection was performed using serological and molecular methods, complemented by morphological blood examinations. Sequence analysis was used to characterize pathogen strains. The findings revealed that *B. canis* DNA was detected exclusively in pet dogs, with a prevalence of 50.0%. *A. phagocytophilum* was identified in both pet and hunting dogs, with prevalence rates of 27.5% and 4.4%, respectively. Although hunting dogs are more frequently exposed to tick habitats, they exhibited lower pathogen prevalence, likely due to the stricter preventive measures implemented by their owners to guard against tick bites. This study highlights the critical role of regular screening and preventive strategies in minimizing the risk of tick-borne diseases among urban canine populations in Lithuania.

## THE ROLE OF THE EUROPEAN PINE MARTEN (*Martes martes*) IN TRANSMITTING SARCOCYSTIS PARASITES

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*Keywords: Sarcocystis, European pine marten, cervids, cox1, molecular identification*

Members of the genus *Sarcocystis* are cyst-forming coccidian parasites infecting mammals, birds, and reptiles. They are characterized by a prey-predator two-host life cycle. Sarcocysts form in the muscles and central nervous system of the intermediate host, while oocysts and sporocysts develop in the small intestine of the definitive host. In the period of 2023–2024, intestinal samples of 20 European pine martens (*Martes martes*) collected in Latvia were examined. *Sarcocystis* spp. were identified by species-specific PCR targeting the *cox1* gene and subsequent sequencing. Under a light microscope, sporocysts of *Sarcocystis* spp. were observed in 14 samples (70%), while using molecular methods, they were detected in 16 animals (80%). Four *Sarcocystis* species were identified in the intestinal samples of European pine marten. The prevalence of the predominant *Sarcocystis* sp., *S. entzerothi* (50%) was significantly higher than that of *S. capracanis* (20%), *S. hjorti* (10%), and *S. truncata* (10%). This is the first time in the world that *S. hjorti* and *S. truncata* have been detected in intestinal samples of European pine marten. The present study provides strong evidence that European pine marten may serve as the definitive host for *Sarcocystis* spp. using goats and cervids as intermediate hosts. Also, the research carried out is important for disclosing the definitive hosts of *Sarcocystis* species under natural conditions.

## THE IDENTIFICATION OF *SARCOCYSTIS* SPECIES IN SMALL INTESTINES OF RACCOON DOGS (*Nyctereutes procyonoides*) FROM LITHUANIA

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**Keywords:** *Sarcocystis*, raccoon dog, light microscopy, molecular identification, *cox1*

*Sarcocystis* parasites infect a diverse range of animals including reptiles, birds and mammals, and have a complex two host prey-predator life cycle consisting of an asexual reproductive stage that ends mainly in muscles of the intermediate host (IH) and a sexual stage that takes place in the intestine of the definitive host (DH). IH are infected by consumption of infected food and/or water. The DH (carnivores) become infected while ingesting meat containing mature sarcocysts. Later, sporocysts are formed and then excreted with faeces. Although sarcocystosis in the DH is typically asymptomatic, it can cause nausea, diarrhoea or weight loss.

Raccoon dogs are invasive animals that carry a number of diseases. As an ecological generalist they have adapted to the urban environment. The shortage of information regarding raccoon dogs as DH of *Sarcocystis* species may be attributed to the lack of in-depth investigation of the animal in the past.

In the period spanning 2019 to 2024, a total of 26 raccoon dog carcasses, comprising those killed on the road and dead of natural causes, were collected in Lithuania. Intestinal scraping samples were obtained and analysed using light microscopy. DNA was extracted and subjected to molecular analysis. A nested PCR, targeting *cox1* gene and subsequent sequencing were conducted to confirm the presence of *Sarcocystis* DNA and identify the specific species involved.

The results of the light microscopy examination indicated that only 50% of the samples were positive for the presence of *Sarcocystis* spp. sporocysts and sporulated oocysts. Conversely, as the molecular analysis is more sensitive, it demonstrated that almost 89% of the samples were positive for the parasites. Molecular analysis revealed the presence of 11 different *Sarcocystis* species, which can be grouped by IH animal species: Cervidae – *S. alces*, *S. capreolicanis*, *S. gracilis*, *S. hjorti*, *S. iberica*, *S. linearis*, *S. morae*, and *S. venatoria*; pig and wild boar – *S. miescheriana*; goat – *S. capracanis*; sheep – *S. tenella*.

A review of the intermediate host groups of *Sarcocystis* species indicates that the majority were from Cervidae animals. This provides a basis for inferring dietary preferences in raccoon dogs.

## **ECTOPARASITE COMMUNITIES OF SMALL MAMMALS ACROSS HABITAT GRADIENTS: FROM NATURAL TO TRANSFORMED**

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*Keywords: small mammals, ectoparasites, habitat, anthropogenic*

Human-driven transformation of natural habitats is a widespread and ongoing process with significant ecological consequences. Anthropogenic habitats have been shown to facilitate and elevate the potential of human infections by agents of zoonotic diseases. Small mammals and their ectoparasites play crucial roles in the lifecycles of numerous veterinary and medically significant diseases. Understanding the ecological dynamics of small mammal ectoparasite communities is essential to successfully predict and mitigate potential zoonotic outbreaks. During this study we investigated ectoparasitic chigger mites (Trombidiformes: Trombiculidae), laelapid mites (Mesostigmata: Laelapidae), ixodid ticks (Ixodida: Ixodidae), blood-sucking lice (Psocodea: Hoplopleuridae, Polyplacidae) and fleas (Siphonaptera: Ceratophyllidae, Ctenophthalmidae, Hystrichopsyllidae) found on small mammals. We analysed the variation in prevalence, intensity, and diversity of ectoparasite communities across different habitat types. Small mammals were trapped in Lithuania from 2017 to 2023 during the warm season. Trappings were held in various open habitats, ranging from natural habitats to areas strongly affected by human activity. A total of 1,924 small mammal specimens representing 14 species were examined. More than 20,000 ectoparasite specimens were collected. In total, 51 ectoparasite species were identified during this study. The overall prevalence of ectoparasite infestations was 82.4%, with an average of 13.1 ectoparasite specimens per infested host. All ectoparasite groups showed different infestation patterns across the habitat transformation range.

## PREVALENCE AND RISK FACTORS OF TOXOPLASMOSIS IN LITHUANIAN HOUND DOGS

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**Keywords:** toxoplasmosis, Lithuanian hound dogs, serological testing, rapid tests, ELISA

Toxoplasmosis, a zoonotic disease caused by the parasite *Toxoplasma gondii*, poses a significant threat to both human and animal health. Canines, including hunting dogs, are susceptible to *T. gondii* infection, which may manifest in various clinical forms. This study investigates the seroprevalence of *T. gondii* infection in Lithuanian Hound dogs, categorizing them into hunting and non-hunting groups. By determining the prevalence of *T. gondii* antibodies in these two groups, the objective of the study further aims to evaluate potential risk factors associated with hunting activity. This research contributes to a deeper understanding of *T. gondii* epidemiology in Lithuanian Hound dogs and its possible implications for public health.

A cross-sectional study was conducted on 90 Lithuanian Hound dogs from different regions of Lithuania from March 2023 to December 2024. The study sample was divided into hunting (n = 44) and non-hunting (n = 46) dogs. Blood samples collected from dogs were immediately centrifuged, and the plasma was separated and stored for serological testing and further analysis. Antibodies against *T. gondii* were detected using two methods: an indirect ELISA test (ID Screen® Toxoplasmosis Indirect Multi-species, ID.vet, France) and an immunochromatographic assay (Toxoplasma IgG/IgM Antibody Rapid Test, Hangzhou Immuno Biotech Co., Ltd.). The findings revealed that 31 dogs (34.4%) tested positive for IgG antibodies, while 17 dogs (18.9%) were IgM-positive according to the rapid test. ELISA confirmed 23 positive cases (25.6%), 19 of which were also detected by the rapid test, demonstrating a significant overlap in results, particularly among hunting dogs. Statistical analysis revealed a strong correlation between the rapid test results and ELISA findings ( $p < 0.05$ ), suggesting that rapid testing is a reliable preliminary screening method for *Toxoplasma* infection. Among the 49 dogs fed raw wild animal meat, 14 (28.6%) were ELISA positive, indicating a strong association between diet and infection status ( $p < 0.05$ ). Further analysis using Cramér's V indicated a moderate strength of association ( $V = 0.35$ ). Logistic regression analysis showed that hunting dogs were approximately three times more likely to test positive for *T. gondii* compared to non-hunting dogs (OR = 3.2, 95% CI: 1.5–6.8).

These results highlight the need for increased awareness and preventive measures regarding toxoplasmosis in Lithuanian Hound dogs, particularly in hunting populations and those consuming raw meat. The findings underscore the importance of regular screening and monitoring for this zoonotic disease, which has implications for public health and canine welfare.

## LIFE IN HIGHLY FRAGMENTED FOREST LANDSCAPES – HOW TO ASSESS THE ACCESSIBILITY OF HABITATS FROM FLYING SQUIRREL *Pteromys volans* PERSPECTIVE

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*Keywords: flying squirrel, CatBoost, minimum planar graph, grains of connectivity*

The habitats of the flying squirrel in Estonia form a system of islands – suitable habitat patches of sufficient size for breeding and smaller habitat patches or stepping stones used for transitions between them, which are isolated from each other in the forest landscape. Individuals move through the diverse forest landscape between these habitat patches which are viable for breeding. However, this landscape often also contains larger open areas that the flying squirrel cannot cross. The more such open areas there are in the forest landscape, the smaller the prospects for flying squirrels to move from one habitat patch to another. The connectivity of habitats is extremely important from the perspective of the species’ mobility and dispersal ability, and it is necessary to be able to assess this and, if necessary, implement measures to improve it.

During this analysis, the connectivity of the landscape was assessed at different levels of mobility. For this purpose, the system consisting of islands and movement corridors was described as a minimum planar graph. The nodes of the graph describe the habitats and stepping stones, and the arcs of the graph describe the movement corridors between them. Depending on the length of the corridors and the mobility of the species, the landscape was divided into connectivity units. One connectivity unit describes an area of the landscape within which the habitats and stepping stones are coherent for the flying squirrel, e.g. considering the daily mobility. The main inputs of the graph analysis were the habitat suitability model and movement resistance map, both with pixel size of 10 × 10 m. CatBoost, a novel machine learning algorithm was used to fit the habitat model. A total of 60 environmental variables were used as predictors, including 6 satellite-derivatives, 36 LiDAR-derivatives. A simplified movement resistance map was generated using two parameters: maximum gap distance resembling the maximum horizontal glide distance (30 m) and minimum forest height resembling the minimum height of forest where individuals are able to move (15 m).

Considering the daily mobility, the connectivity of habitats reveals a rather high level of isolation. If the height of the corridor to be traversed is considered to be a minimum of 15 m, then the habitat network of the study area is divided into 632 units and the network of habitats potentially accessible to the flying squirrel is divided into 338 units.

## INVESTIGATION OF *Trichinella* spp. IN PREDATORY MAMMALS FROM LITHUANIA

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*Keywords: Trichinella, molecular identification, wild animals*

Parasites of the genus *Trichinella* cause zoonotic parasitic disease – trichinellosis. These hazardous parasites can be transmitted through the foodborne route in carnivores, omnivores, and scavengers. Previous comprehensive surveys of *Trichinella* parasites in Lithuanian wild predators were carried out 13 years ago. This study aimed to determine the prevalence and intensity of infection and to identify *Trichinella* species in predatory mammals from Lithuania.

In this study, 150 samples of wild predatory mammals were analyzed. The active method of artificial digestion has been used to study the prevalence and intensity of *Trichinella* infection in tested samples. *Trichinella* species were identified using multiplex-PCR.

Of the 150 predatory mammals tested, 33 (22.00%) were positive for *Trichinella* spp. The prevalence of *Trichinella* nematodes was 4.07 times higher in canids than in mustelids. Overall, 330 larvae were isolated for molecular identification of *Trichinella* species. Of the 330 larvae, 96.97% were identified as *T. britovi* and 3.03% were identified as *T. spiralis*. *Trichinella britovi* was found in six out of seven (85.71%) species of carnivorous mammals, badger, pine marten, lynx, grey wolf, red fox, and raccoon dog. *Trichinella spiralis* was detected in one species (14.29%) – American mink. In conclusion, *T. britovi* was the dominant *Trichinella* species in predatory mammals from Lithuania. No mixed *Trichinella* species infections were found in the animals analyzed.

## MOLECULAR DETECTION OF VECTOR-BORNE PATHOGENS IN RED SQUIRRELS (*Sciurus vulgaris*) IN LITHUANIA

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**Keywords:** red squirrel, *Sciurus vulgaris*, *Borrelia burgdorferi*, Lyme disease

Lyme disease, caused by bacteria *Borrelia burgdorferi* sensu lato, is the most common vector-borne disease in North America and Europe. Over the past few decades, the number of cases has been increasing, partly due to expanded migration of ticks and their hosts. Understanding whether *Sciurus vulgaris* (red squirrel) might serve as a significant reservoir host for *Borrelia burgdorferi* s.l. is crucial, as this wild rodent frequently inhabits peri-urban and urban areas and is regularly exposed to ticks while moving on the ground. This study aimed to evaluate the potential role of red squirrels in the maintenance and transmission of *Borrelia burgdorferi* s.l. in urban areas by analyzing samples from red squirrels collected in various parks within Kaunas city, Lithuania. A total of 45 red squirrel tissue samples were analyzed for the presence of *Borrelia* DNA using real-time PCR with primers specific to the *Borrelia* genus. Positive samples were further analyzed by PCR amplification of the ospA gene, intergenic spacer (IGS) and fla regions, followed by sequencing of the amplified products. Real-time PCR analysis revealed that the *Borrelia* pathogen was detected in 37.7% of the analyzed squirrel samples. Detection rates of *Borrelia* were 17.7% for the ospA gene, 20.0% – for the IGS region and 37.7% – for the fla gene. Sequencing revealed the presence of three pathogenic species, *Borrelia afzelii*, *Borrelia garinii* and *Borrelia burgdorferi* sensu stricto, which are known to cause Lyme disease. These findings suggest that red squirrels may play a significant role in the ecology of Lyme disease within urban environments by serving as a reservoir for *Borrelia* spp. This highlights the importance of monitoring wildlife and vector populations in tick-endemic regions to better understand disease dynamics and support the development of effective public health interventions.

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## **Dictyocaulus spp. LUNGWORMS IN WILD UNGULATES IN LITHUANIA**

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*Keywords: Dictyocaulus skrjabini, Dictyocaulus cervi, Dictyocaulus capreoli, Dictyocaulus viviparus bisontis, Lithuania*

This study aimed to identify the species of lungworms present in red deer *Cervus elaphus*, roe deer *Capreolus capreolus*, moose *Alces alces*, and European bison *Bison bonasus* using both morphological and molecular methods. Between 2021 and 2024, lung samples were collected and analyzed from 41 red deer, 33 roe deer, 14 moose, and 31 European bison to detect the presence of lungworms. The parasites were extracted from the trachea, bronchi, and bronchioles, with genus-level identification performed based on morphological characteristics. DNA was isolated from adult lungworms using the QIAGEN QIAamp DNA Mini Kit, and the *cox1*, *18S SSU*, and *cytB* genes were amplified by PCR and sequenced. In total, 1,122 lungworms were collected, distributed as follows: 1,034 from red deer, 29 from roe deer, 49 from moose, and 10 from European bison.

The morphological characteristics of the lungworms indicated that the ungulates were infested with *Dictyocaulus* spp. Furthermore, phylogenetic analysis identified three prevalent *Dictyocaulus* species in cervids and one dominant species in European bison. This study highlights the critical role of molecular analysis in accurately identifying *Dictyocaulus* species, particularly in wild ungulates and domestic ruminants.

## BAT MIGRATION ALONG THE NORTHERN COAST OF THE NORTH SEA

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*Keywords: bats, migration, coastline, Motus*

Several bat species in the boreal region undertake short- or long-distance migrations between their summer roosts and winter hibernation sites. Long-distance migrants are at greater risk due to the rapid development of both onshore and offshore wind energy, as their migration routes often overlap with areas of high importance in wind energy development. Predicting the cumulative effects of energy production along bat migration routes is very challenging and extends beyond national borders.

We study bat spatiotemporal activity along the southwest coast of Norway, with special attention to determining the activity patterns of migratory species like the Nathusius's pipistrelle (*Pipistrellus nathusii*). Our aim is to collect comprehensive data on bat community composition and activity in a large proportion of the coast. In the upcoming years, we also plan to capture and equip bats with radio transmitters. We will deploy Motus towers and utilize the Motus wildlife tracking network around the North Sea to track movements of individual bats in greater detail.

As part of an ongoing study on bat migration, we deployed 50 bat acoustic detectors along the coastline and nearby lakes in the summer and autumn of 2024. The study area spans from Utsira island to Lista peninsula in the south, covering ca two hundred kilometres in a straight line.

The poster presentation will focus on the preliminary findings on species composition and timing of the autumn migration along the Norwegian coastline. Little is known of the species-specific timings of peak migration in Norway. Therefore, site-specific differences between detector locations will allow us to analyze patterns not only in the timing of migration, but also the impacts of environmental factors such as landscape composition and configuration, and weather conditions.

## MONITORING OF EURASIAN OTTERS (*Lutra lutra*) IN LATVIA: OCCURRENCE, DISTRIBUTION AND POPULATION TRENDS

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*Keywords: Lutra lutra, distribution, occurrence, population, monitoring*

According to the applied monitoring methodology, distribution is the most accurately interpretable indicator of the otter population status. The number and location of otter occurrence signs at inspection points are used as the main variables for characterizing population changes or stability. As long as the occurrence and abundance of the signs do not change significantly, the population can be considered as stable.

Since 2014, the monitoring of otters in Latvia has been conducted twice (in 2014–2017 and in 2020–2023). A grid of 10 × 10 km squares is used for assessing otter occurrence and four preliminary selected sites at banks and shores of different water bodies in each square are visited to check for the presence of otter signs. In total, 652 squares were checked. Primarily, the sites where roads cross the watercourses were chosen because such places are easily accessed and convenient to survey. Bridges and culverts are of particular attention since the signs at these constructions are inspected first. Indirect evidence of otter presence was used, including the number of fresh signs (footprints, excrements and territorial marks) and the number of older excrements. Data from 2020 to 2023 were compared with the previous monitoring results in 2014–2017. In total 2,676 sites were inspected, 94% of which were associated with bridges and culverts. The remaining 6% of sites were on the seacoast and the shores of stagnant inland water bodies.

The presence of otters was found in 648 squares, in total at 2,159 locations (80.7%). Although the proportion of sites inhabited by otters was slightly lower than in the previous monitoring cycle (650 inhabited squares with 84.1% of the tested sites inhabited), the differences were not statistically significant ( $\chi^2 = 20,000$ ,  $p = 0.22$ ). The differences in the results may be caused by extensive road reparation and bridge reconstruction work, which made it impossible to access or find the otter signs. Thick vegetation along the banks of water bodies also made it difficult to find the signs of otters.

The monitoring results indicate that otters are widespread and found throughout the territory of Latvia. However, compared to the previous monitoring cycle, in 2023, there was a decrease in fully inhabited squares in the SW part of Latvia and along the eastern coast of the Gulf of Riga. The occurrence of otters in different water bodies depends on their type. Comparing the sea coast, watercourses and stagnant water bodies, otters were found slightly less frequently near inland stagnant water bodies. It can be concluded that the otter population is not fragmented and 3,500 to 4,000 otters are still living in Latvia, similarly as estimated at the end of the 20<sup>th</sup> century.

## INFECTIONS IN WILD CARNIVORES BY *Babesia* sp., *Anaplasma* sp., AND *Mycoplasma* spp.

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*Keywords: wild carnivores, Babesia sp., Anaplasma sp., Mycoplasma spp.*

The aim of this research was to investigate the prevalence and genetic diversity of *Babesia* sp., *Anaplasma* sp., and *Mycoplasma* spp. infections in wild carnivores, contributing to a better understanding of their role as reservoirs for zoonotic pathogens and the dynamics of vector-borne disease transmission in wildlife populations.

Seventy spleen samples from wolves (*Canis lupus*) and four samples from golden jackals (*Canis aureus*) were collected between 2018 and 2024 in Lithuania and stored frozen until analysis. Genomic DNA was extracted from the spleen tissues of wild carnivores using the Genomic DNA Purification Kit (Thermo Fisher Scientific, Vilnius, Lithuania), following the manufacturer's recommendations. The presence of protozoan and bacterial pathogens was screened using conventional and nested PCR techniques, with positive and negative controls included in each PCR run. PCR products were visualized by electrophoresis on a 1.5% agarose gel. Positive PCR products were extracted from the gel and purified using the GeneJET™ Gel Extraction Kit (Thermo Fisher Scientific, Vilnius, Lithuania).

Partial sequences of *Babesia* sp., *Anaplasma* sp., and *Mycoplasma* spp. were analyzed using the MegaXI software package and aligned with previously uploaded sequences in GenBank via BLASTn. Phylogenetic trees were constructed using the maximum-likelihood method with the Tamura-Nei model.

The study showed that *Babesia canis* was the most widespread pathogen, detected in 38% of the tested individuals. *Anaplasma phagocytophilum* was identified in 12% of the samples, while *Mycoplasma* spp. infections were found in 7% of the tested carnivores. These results highlight the significant role of wild carnivores as potential reservoirs for zoonotic pathogens, with important implications for wildlife conservation and public health strategies.

## NON-INVASIVE GENETIC ANALYSIS OF THE LATVIAN BROWN BEAR POPULATION

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*Keywords: Ursus arctos, genetic monitoring, microsatellites, diversity, population structure*

The genetic monitoring of wild populations can provide valuable information on demographic and other processes, as well as direct information about changes in genetic diversity. Genetic diversity is crucial for the long-term survival and adaptation of populations, and is the foundation of the other levels of biodiversity – species and ecosystem diversity.

The brown bear population in Latvia has been increasing in recent years, and monitoring of the population has been carried out since 2015. Genetic monitoring of the population was initiated in 2018, using non-invasively collected samples. The sample types collected for DNA extraction include hair, scats, footprints, and saliva. Samples are collected during bear monitoring activities, and hair samples are also specifically collected in hair traps constructed using barbed wire. Samples are also collected from sites where bears have destroyed bee hives as well as abandoned apple orchards where bears are feeding in the autumn. DNA is extracted from samples using the QIAamp DNA Micro Kit (Qiagen) or the E.Z.N.A Stool DNA Kit (Omega Bio-tek). If the sample species is unclear (usually for hair samples), DNA is tested using a bear-specific marker prior to further genotyping. Genotyping is done with 12 microsatellite markers, as well as a sex determination marker.

From samples collected between 2018 and 2023, 83 individuals (49 males, 17 females, 17 with sex not determined) have been genetically identified. These genetic analyses enable determination of the sex of an individual, as well as monitoring of the movement of individuals. The development of a database of bear genotypes also enable the determination of genetic diversity, as well as population structure and pedigree analyses. It is also planned to genotype Estonian bears, particularly from the areas bordering Latvia, to investigate the movement of individuals from the relatively dense local population in Estonia into Latvia.

## COMPARATIVE ANALYSIS OF INTRASPECIFIC AND INTERSPECIFIC GENETIC VARIABILITY IN *Pipistrellus* SPECIES USING CYTB AND ND1 MARKERS

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*Keywords: Pipistrellus, cytB, ND1*

The genus *Pipistrellus*, prominently featuring species such as *Pipistrellus nathusii*, *P. pygmaeus*, and *P. pipistrellus*, is widespread across Lithuania and is integral to the local ecosystem due to its role in controlling pest populations. These bats are also significant from an epidemiological perspective as they harbor various pathogens with zoonotic potential, which can pose health risks to other animals and humans. Recent global health crises, including the COVID-19 pandemic, have heightened interest in bats, emphasizing the need to enhance our understanding of their genetic composition. Studying intraspecific and interspecific genetic variability is essential not only for conservation and management of bat populations but also for better understanding the dynamics of disease transmission. Current morphological methods of species identification are often inadequate due to the subtle differences among species and individuals, highlighting the necessity for precise molecular techniques in bat research. This investigation employs mitochondrial DNA markers, specifically cytochrome b (cytB) and NADH dehydrogenase subunit 1 (ND1), to assess both intraspecific and interspecific genetic variability within the *Pipistrellus* genus. Bat individuals representing the three species within this genus, were sampled from a variety of geographic locations across Lithuania. Genomic DNA was isolated from either blood samples or internal organs such as the spleen, heart, and lungs. Extensive sequence analyses were conducted using DnaSP software to quantify variable sites, haplotype diversity, and nucleotide diversity, offering detailed insights into the genetic structure of these populations.

## SMALL MAMMAL AGROECOLOGY IN LITHUANIA IN THE CONTEXT OF PRESERVING BIODIVERSITY

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*Keywords: commercial orchards, abundance, agricultural intensity, rodent diversity*

This study, focused on commercial orchards and berry plantations in Lithuania, evaluated the diversity, relative abundance, population structure, and ecological characteristics of small mammal species, also considering their broader implications in a global context both as pests and ecosystem service providers. Thirteen small mammal species were captured during the study in 2018–2024, with the most abundant being the common vole (*Microtus arvalis*), yellow-necked mouse (*Apodemus flavicollis*), striped field mouse (*A. agrarius*), and bank vole (*Clethrionomys glareolus*). Their relative abundance and proportion varied according to year, season, and habitat, with no significant population outbreaks. Seasonal trends, habitat complexity, and orchard management practices strongly influenced the dynamics of these species. In other countries, proximity to semi-natural habitats was linked to increased species richness and population stability, underscoring the importance of landscape structure. Trophic resource partitioning, as revealed through stable isotope analyses, showed niche differentiation among species, allowing for coexistence and reducing competition. *Clethrionomys glareolus* in apple orchards exhibited an omnivorous diet, with its trophic niche closer to insectivores than granivores and herbivores. *Sorex* shrews were underrepresented, likely due to pesticide use and prey scarcity. Agricultural intensity emerged as a key driver of small mammal abundance and diversity. Frequent mowing, soil tillage, and pesticide or rodenticide application reduced their populations, while less intensive or sustainable approaches created favourable conditions for biodiversity. Elevated levels of elements such as Cu, Mn, Co, and Pb in the muscle and bone tissues of small mammals in commercial gardens highlighted the impact of fertilizer and pesticide use. Detection of *Sarcocystis* pathogens in small mammals emphasizes the need to consider health implications in these populations. Concluding, Lithuanian orchards demonstrated a significant contribution to biodiversity within agricultural landscapes. Reviews of small mammal diversity across decades suggest that major shifts in land use and climate have impacted community compositions, with potential implications for orchard agroecosystems. Our findings underscore the importance of adapting agricultural practices to maintain biodiversity and functional small mammal roles in orchards, highlighting key areas for future research.

## **WOLF (*Canis lupus* L.) POPULATION AGE AND SEX STRUCTURE IN LITHUANIA**

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*Keywords: males, females, hunting season, canine tooth*

Long-term observations of the wolf population state reveal that it is growing in the territory of Lithuania. The last decline in the number of wolves was recorded in the first decade of this century. Therefore, starting from the 2005–2006 hunting season, a quota for the number of wolves allowed to be hunted was set. At the beginning of the limitation, the number of wolves allowed to be hunted was cautiously, therefore it was small. Since then, the wolf population has been growing. The hunting limit has also been increased. Over three hundred wolves were allowed to be hunted in the last hunting season.

Starting from the 2018–2019 hunting season, hunters who have hunted a wolf, are required to record basic information about the hunted wolf (location, time of hunt, sex of the wolf, scabies mite infestation, external injuries) and submit samples for testing (muscle for genetic testing, canine tooth for age testing, uterus for reproductive testing). The exact age of each hunted wolf is determined based on the annual cementum layers at the root of the canine tooth. 1,146 wolves were hunted between the 2018–2019 and 2023–2024 hunting seasons. Samples were collected from 98.3% of the hunted wolves.

After examining the canines of wolves hunted during the 2023–2024 hunting season, it was found that 51.9% of individuals were 5–9-month-old cubs. This indicator has fluctuated from 48.3% to 60.8% over the past six hunting seasons. In the age group of first-year cubs (wolves up to a year old), the proportion of males and females was 1:0.72. The sex distribution between hunted males and females (in all age groups) during this period varied from 1:0.90 to 1:0.69. In older age groups, fewer wolves were hunted. For example, second-year cubs (wolves aged 1 to 2 years) accounted for an average of 7.0% of all wolves hunted in Lithuania during recent seasons. The oldest hunted wolves (two males) were in their thirteenth year. They were hunted during the 2023–2024 hunting season. These were the oldest wolves that have been hunted in the past six seasons. In previous seasons, the oldest wolves hunted were in their tenth and twelfth years.

In Lithuania, the average age of hunted adult wolves (excluding first- and second-year cubs) during the past six hunting seasons ranged from 4.85 (2018–2019 season) to 5.84 (2023–2024 season) years. Since the 2020–2021 hunting season, this indicator has been increasing (from 4.12 to 5.84 years).

## DOMESTIC DOG AS A POTENTIAL SPREADER OF *Sarcocystis* PARASITES IN VILNIUS CITY

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Sarcocystosis is a parasitic infection caused by protozoans of the *Sarcocystis* genus, which rely on a two-host life cycle involving intermediate and definitive hosts. Intermediate hosts develop sarcocysts in their muscles following ingestion of contaminated food or water, while sporocysts form in the intestines of definitive hosts where they complete their life cycle. Domestic dogs are an essential part of human and animal life, forming inseparable bonds with people in cultures around the world. However, alongside their role as companions, they are also carriers of numerous parasitic infections, including *Toxocara spp.*, *Echinococcus spp.*, *Dipylidium spp.*, *Ancylostoma spp.* and *Sarcocystis*. Even though there are studies related to *Sarcocystis* spp. presence in the feces of wild canids, data on the presence of these parasites in the feces of domestic dogs, especially in Europe, remains scarce. Thus, this study aimed to examine the prevalence and diversity of *Sarcocystis* species in dog feces collected in Vilnius, Lithuania.

A total of 25 fecal samples from domestic dogs were collected from urban areas in Vilnius. Samples were processed using flotation-sedimentation methods. Microscopic examination revealed no visible sporocysts or oocysts. However, molecular analysis utilizing nested PCR and sequencing of mitochondrial *cox1* biomarker revealed the overall prevalence of *Sarcocystis* spp. in the samples to be 32%. Additionally, four *Sarcocystis* species were identified to be present in the feces, such as *S. tenella* (32%), *S. miescheriana* (12%), *S. hjorti* (8%), and *S. arieticanis* (4%). These findings indicate that intermediate hosts such as sheep, pigs, and deer may contribute to the observed distribution patterns.

For the first time molecular methods were employed to analyse *Sarcocystis* diversity in the feces of domestic dog collected in Lithuania. Results of the study suggests that dietary and environmental factors, including exposure to raw or undercooked meat, may facilitate the spread of *Sarcocystis* in domestic dogs. Additionally, this study advances our understanding of their epidemiology and guides future research and control initiatives across Europe and beyond.

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