Reg. No: 2024/53/B/NZ8/02682; Principal Investigator: dr Danijela Popović

Reconstruction of the evolution history of brown bear in the Balkans

The brown bear (*Ursus arctos* L.) is a magnificent and iconic species that survived the massive extinction event that wiped out many large animals during the Late Pleistocene epoch. Currently, it is one of the most widely distributed bear species inhabiting Eurasia and North America, and it is the only bear species that roams continental Europe. Historically, this large carnivore has been widespread throughout Europe, but human activities, such as hunting and habitat destruction, have greatly reduced their numbers and territories. Currently, they are mostly confined to fragmented landscapes that are heavily influenced by human presence. While some European countries have stable brown bear populations, others are at risk of local extinction. Recent estimates indicate that Europe is home to approximately 17,000 brown bears, clustered into ten subpopulations. The Balkans are home to three of them: the Dinaric-Pindos, Carpathian, and Eastern Balkan populations, with contact zones in Serbia.

The goal of our project is to uncover the evolutionary history of brown bears in the Balkans over the past 50,000 years using advanced biomolecular techniques, such as whole-genome sequencing and stable isotope analysis. Preliminary results of Holocene brown bears in Serbia have revealed rich diversity in mitochondrial DNA (mtDNA), including all known European lineages and a newly discovered one, indicating that brown bears were more genetically diverse in the past. However, genetic information about brown bears in the Balkans is still limited and mainly consists of short mtDNA fragments.

Our project will analyse approximately 100 ancient and 50 modern brown bear samples. The ancient samples come from bones found in palaeontological and archaeological sites in Serbia, Bulgaria, Romania, and Italy, dating from the Late Pleistocene to the Middle Ages. Modern samples will be obtained from the Dinaric-Pindos population by analysing non-invasive probes collected in recent years and from tissue samples of animals that died accidentally or were hunted.

We will use the latest techniques in genomics, paleogenomics, and isotope biochemistry. By studying both mitochondrial and nuclear DNA, we will gather genome-wide data for conducting temporal and spatial analyses. The genomic data will be combined with the nitrogen and carbon composition of bone collagen to infer the feeding habits of brown bears and understand their dietary flexibility. The direct radiocarbon dates of the bone samples will serve as the baseline for the timescale of the data obtained. This will help us understand the demographics, ancestry, migrations, relationships between individuals and populations, and other factors, such as genomic erosion through time, selection signatures, and adaptations to changing living conditions. We expect that this project will bring us closer to the answer as to why brown bears survived while cave bears became extinct.

This project will provide new insights into the history of brown bear in the Balkans, which will be crucial for conservation policies, both regionally and across Europe. Genomic data spanning the last 50,000 years, combined with an understanding of past and current environmental and human pressures, will be invaluable for ensuring the future of these animals.