From genotype to ecotype in European roe deer

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As a widespread ungulate, European roe deer (*Capreolus capreolus*) is exposed to various environmental variables across its range and is recognized as one of the most adaptable species among cervids due to its ability to withstand significant anthropogenic pressure and thrive in landscapes impacted by human activity. The existence of two mtDNA lineages in this species—the European and the Siberian (an introgression of *C. pygargus* mtDNA into *C. capreolus*)—was demonstrated based on an analysis of the mitochondrial control region. The Siberian lineage is the most frequent in the eastern part of the continent and declines toward Central Europe. The European lineage consists of three clades (Central, Eastern, and Western), each composed of several haplogroups.

In our study, which primarily focuses on the genetic background of roe deer ecotypes, we estimated the genetic structure of roe deer based on the whole mitochondrial genome. We employed whole-genome sequencing at 15-fold depth for 40 roe deer samples from Slovenia and Poland. Additionally, we used publicly available mitogenomes. The MitoZ tool was used for mitogenome assembly and annotation, followed by multiple sequence alignment using MAFFT and network construction. To study genetic differentiation between ecotypes, we analysed approximately 1 million unlinked nuclear single-nucleotide polymorphisms (SNPs) from the nuclear genome to construct a hierarchical clustering tree and perform multidimensional scaling (MDS) plotting. Our analysis confirmed the presence of the three mitochondrial clades of roe deer in Europe. More complex structure was revealed by analysing SNP data across the roe deer ecotypes.