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First ancient DNA sequences from the Late Pleistocene red deer (Cervus elaphus) in the Crimea, Ukraine

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The Late Pleistocene has been a period of significant population and species turnover and extinctions among the large mammal fauna. Massive climatic and environmental changes during Pleistocene significantly influenced the distribution and also genetic diversity of plants and animals. The model of glacial refugia and habitat contraction to southern peninsulas in Europe as areas for the survival of temperate animal species during unfavourable Pleistocene glaciations is at present widely accepted. However, both molecular data and the fossil record indicate the presence of northern and perhaps north-eastern refugia in Europe. In recent years, much new palaeontological data have been obtained in the Crimean Peninsula, Ukraine, following extensive investigations. The red deer (Cervus elaphus) samples for aDNA studies were collected in Emine-Bair-Khosar Cave, situated on the north edge of Lower Plateau of the Chatyrdag Massif (Crimean Mountains). The cave is a vertical shaft, which functioned as a huge mega-trap over a long period of time (probably most of the Pleistocene). The bone assemblages provided about 5000 bones belonging to more than 40 species. The C. elaphus bones were collected from three different stratigraphical levels, radiocarbon dated by accelerator mass spectrometry (AMS) method. The bone fragments of four specimens of red deer were used for the DNA isolation and analysis. The mtDNA (Cytochome b) was successfully isolated from three bone fragments and the cytochrome b sequences were amplified by multiplex PCR. The sequences obtained so far allowed for the reconstruction of only preliminary phylogenetic trees. A fragment of metatarsus from level dated to ca. 48,500±2,000 years BP, yielded a sequence of 513 bp, allowing to locate the specimen on the phylogenetic tree within modern C. elaphus specimens from southern and middle Europe. The second bone fragment, a fragment of mandible, collected from level dated approximately to ca. 33,500±400 years BP, yielded a sequence (696 bp) locating this specimen much closer to the modern C. elaphus specimens from China and Far East. From the third bone fragment (metatarsus), dated between ca. 12,000 years BP and 30,000 years BP, the sequence of only 346 bp has been obtained. It locates this specimen between European and Asiatic haplogroups. The preliminary results of analysis of the DNA from Crimean C. elaphus fossils reveal the great genetic heterogeneity and a complex phylogeographical pattern of the material studied. The obtained results support the opinion that Crimean Peninsula was the most north-eastern refugium in Europe during Late Pleistocene playing a major role in recolonization and dispersal processes of temperate species during and after the Late Pleistocene in this part of the Euro-Asian continent.