



CHARACTERIZING PLEISTOCENE CAVE BEAR *URSUS SPELAEUS* ROSENMÜLLER BASED ON ENAMEL PROTEOMICS

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This study presents the proteomic characterization of the cave bear species (*Ursus spelaeus* Rosenmüller, 1794) based on the dental enamel proteome from the upper Pleistocene period at Cova Eirós, an archaeological and palaeontological site, located in the Sierra de Ouribio, Lugo (Galicia, Spain). It is known for its significant cave bear population, representing the westernmost distribution of this species. A total of 9 dental enamel samples from upper and lower first and second molars were analyzed. Peptides were recovered from the fossil material using tandem mass spectrometry (MS/MS) technology. For their identification, a reference database was created. The sequences were obtained from open access databases and belong to three species of the genus *Ursus*: *U. americanus*, *U. maritimus*, and *U. arctos*, and one species of the genus *Ailuropoda*, *A. melanoleuca*. Additionally, proteins of potential interest were mapped and predicted from previously published *U. thibetanus* genome, for inclusion in the comparative database. Due to the optimal state of conservation, it has been possible to successfully extract peptides from all the cave bear samples, which has allowed us to identify and sequence the dental proteome of this species for the first time. Following a comparison of the proteomes, the results reveal that this upper Pleistocene ursids differs at a proteomic level from other representatives of the genus *Ursus*, which includes some extant bear species. Consistent variations at the amino acid level in protein sequences suggest these differences, enabling a preliminary phylogenetic analysis through protein sequences. This highlights the potential of the proteome in studying the phylogeny of this emblematic species. This study presents a novel application of palaeoproteomics in Pleistocene cave bears, offering a rich valuable information about their evolutionary relationships and even their biological sex.

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