PALAEOENVIRONMENTAL RECONSTRUCTION BASED ON DENTAL WEAR AND STABLE ISOTOPE ANALYSIS OF CERVIDS

THESES OF THE DISSERTATION

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Introduction

Cervids were and are abundantly present in the Carpathian Basin throughout the entire Pleistocene and Holocene periods. Their remains are among the most common findings from Quaternary large mammal bearing localities. The members of the group were present in both glacials and interglacials. During the aformentioned period, the species of six genera (*Alces*, *Capreolus*, *Cervus*, *Dama*, *Megaloceros*, *Rangifer*) were present within the studied area. Their widespread distribution and wide environmental tolerance make them an ideal group for palaeoenvironmental analyses. Their constant presence ensures that their remains are suitable for taxon-free analyses such as dental wear and stable isotope analyses throughout the period, on which this study focuses.

The dissertation can be divided into two main parts: (1) the re-evaluation of the dental microwear method based on the examination of extant ungulate specimens; (2) as well as palaeoenvironmental examination of the Quaternary of Hungary based on cervid dental remains.

New results in the field of dental microwear analysis

In the first part of the dissertation, the possibilities of removing some methodological limitations of the low-magnification dental microwear method (LM-method; SOLOUNIAS & SEMPREBON, 2002; SEMPREBON ET AL., 2004) were examined. This was essential to ensure that the largest possible number of samples could be studied from the – often incomplete and fragmentary – Quaternary cervid materials during the second part of the dissertation. To examine these possibilities, the dentitions of extant ungulates (roe deer, red deer, domestic sheep and west-caucasian tur) from the Mammalia Collection of the Hungarian Natural History Museum were examined. High definition epoxy replicas were made from the examined dental elements, and the observable enamel scars were then quantified on them. My study focused on the following issues: I.) Are the results of independent observers directly comparable with each other, with similar environmental implications? II.) Is the microwear pattern homogenous on the whole enamel surface of the second upper and lower molars (M2/m2)? III.) Are other tooth positions suitable for LM microwear analyses? IV.) On how many 0.16 mm^2 areas is it necessary to quantify the microwear pattern to sufficiently represent the microwear of a given tooth?

To answer issue **I.)**,the microwear pattern was examined on two predefined areas on the replicas of all right molars and premolars of eight specimens (196 areas in total) by an independent researcher (Dr. Attila Virág) and by myself. The data of the two observers were then compared with each other, intraclass correlation coefficients (ICC) were calculated and standard major axis regressions were fitted on them.

For examining issues **II.) and IV.),** the microwear pattern was recorded on 1752 areas on the replicas of all right molars and premolars of six specimens, to explore the background distribution of the microwear pattern on different tooth positions. The paracone and protoconid areas of the M2/m2 teeth were compared to other occlusal areas of these tooth positions by two-sided t-tests, and their descriptive statistics were also checked. To check the variance of the microwear pattern between different teeth, the descriptive statistics (mean, median, SD, range, skewness, kurtosis) of each individual tooth was examined and they were compared to the results obtained on M2/m2 with ANOVA. For determining the required number of sampling areas per tooth, the average of increasing number of sampling areas were summarized and averaged until the resulting value sufficiently approached the mean and fell within the SD range of the microwear background distribution of the same tooth.

Based on the results of the aformentioned examinations, I drew the following conclusions:

I. Based on the results of the two independent observers, the robustness of the LM-method was further established. By comparing the results of the independent observers, high intraclass ($ICC_{scratch}$=0.84; ICC_{pit}=0.94) and correlation coefficient values ($r^2_{scratch}$=0.57; r^2_{pit}=0.80) were calculated for both the scratch and the pit numbers (**SZABÓ & VIRÁG, 2021**).

II. It was proven that the m2 protoconid and M2 paracone areas, traditionally used for LM-studies (MERCERON ET AL., 2004; RIVALS ET AL., 2009), show similar microwear as other enamel areas of the same tooth. Thus the LM-analysis can be executed on any part of these teeth, it is not necessary to confine the method to the protoconid and paracone areas (**SZABÓ & VIRÁG, 2021**).

III. It was proven that, apart from the originally determined M2/m2 positions, other tooth positions are also suitable for LM-analysis. Excluding the first two premolars, all other premolars and molars have statistically similar microwear as that of the M2/m2 positions (**SZABÓ & VIRÁG, 2021**).

IV. By comparing the microwear indices (no. of scratches/no. of pits) of the different tooth positions, it was further proven that, apart from the first two premolars, all other molars are suitable for LM analysis. The wear indices of the molars and premolars – excluding the first two premolars – were similar to the indices of the second molars (**SZABÓ & VIRÁG, 2021**).

V. It was proven that it is possible to characterize the microwear background distribution of any given tooth by examining five randomly selected 0.16 mm^2 sampling areas. The average of five areas falls close to the typical microwear signal of the entire occlusal surface of the same tooth (**SZABÓ & VIRÁG, 2021**).

These conclusions are especially important if palaeoenvironmental studies can only utilise fragmentary dental materials. These results ensure that it is possible to expand the LM-method, and by this expansion increase the number of potential samples available for environmental studies. Furthermore, the minimal amount of work was determined to ensure robust, comparable results and relatively fast sample procession.

Palaeoenvironmental reconstruction of the Carpathian Basin

In the second part of the dissertation, the Quaternary climatic shifts of the Carpathian Basin were examined. Cervids were selected for this analysis, because their representatives were continuously present during the examined period, and their dental elements are frequently found in Pleistocene and Holocene localities. The following studies can be divided into two main topics: dental wear analysis and stable isotope analysis.

From the different dental wear methods, the mesowear (MIHLBACHLER ET AL., 2011) and the LM-microwear (SOLOUNIAS & SEMPREBON, 2002; SEMPREBON ET AL., 2004) analyses were conducted on the selected specimens.

The mesowear analysis – representing the abrasive properties of the food items consumed by the animals during their entire life – was conducted by the methodology of MIHLBACHLER ET AL. (2011). The wear of the coni was evaluated based on photographs taken from the selected dental elements, excluding unworn and fully eroded ones. The teeth were classified into seven groups, ranging from zero to six. This scale represents the ever-increasing intake of highly abrasive plant materials. This analysis was conducted on 727 molars and premolars.

The results of microwear analysis represent the consumed plant materials of the last few days of an animal’s life. High definition epoxy replicas were made from the enamel surfaces of the selected specimens (in total 73 dental elements). Then the microwear pattern was quantified on these replicas. If possible, five 0.16 mm^2 areas were checked on each replica to ensure that their average represents the microwear pattern of the whole tooth. Based on the results, the specimens were classified into three dietary categories: browser, mixed feeder, or grazer.

For the stable isotope analyses, the dental elements of in total 31 specimens were selected from nine localities. Bulk enamel samples from the thoroughly cleaned teeth were taken with tungsten-carbide coated drill bits. Each sample represent the whole time frame of the enamel forming, roughly 1-1.5 years. The isotopic measurements were performed in the Isotope Climatology and Environmental Research Centre, Atomki, Debrecen. Then I performed the environmental evaluation of the structural phosphate ($\delta^{18}O_{PO4}$) and carbonate ($\delta^{13}C_{CO3}$).

Based on the stable oxygen analysis of the dental enamel, it is possible to gain insight of the isotopic composition of precipitation, and consequently of the average temperature of past times (DANSGAARD, 1964; LONGINELLI, 1984). To uncover the connection between the isotopic composition of the enamel and the average temperature, a regional temperature-precipitation $\delta^{18}O$ regression was calculated for the Carpathian Basin based on data from seven regional GNIP (Global Network of Isotopes in Precipitation) laboratories. Furthermore, based on data obtained from literature, a regression was calculated to estimate the connection between the bone and enamel $\delta^{18}O$ values of extant cervids (and their close relatives) and the isotopic values of the water consumed by these specimens. The results of these regressions allow to make palaeotemperatures esimates from the $\delta^{18}O_{PO4}$ values of cervid enamel samples.

By utilising the stable carbon isotopic measurements, it is possible to check if the selected specimens were diagenetically altered or not (IACUMIN ET AL., 1996; PELLEGRINI ET AL., 2011). Furthermore, based on the results of such analyses, it is possibble to gather information on the past vegetation. Plants utilising different photosynthetic pathways have differring carbon isotopic print (O'LEARY, 1981; KEELEY & RUNDEL, 2003), thus, by studying the animals that consume these plants, it is possible to draw conclusions regarding the closedness of the surrounding vegetation and the photosynthetic pathway used by the plants (CERLING & HARRIS, 1999; BONAFINI ET AL., 2013).

Based on the aformentioned examinations, I drew the following conclusions:

VI. Based on the mesowear analysis of 727 Pleistocene and Holocene cervids, it was concluded that Early-Pleistocene specimens had lower average mesowear score, than cervids from the Late-Pleistocene. This change could be the result of the vegetation becoming more open from the Early-Pleistocene onwards (**SZABÓ ET AL., 2021**).

VII. It was concluded that mesowear scores of specimens from glacial periods are relatively high. Such high values represent a grazer or graze dominated mixed feeder diet. The microwear signal from the glacial periods in most cases also suggest a grazer diet for the animals that lived during these intervals. Such wear pattern suggests an open, steppe-like vegetation in the Carpathian Basin for these cooler periods. The area – apart from some extreme exceptions – could have had some minimal tree cover during these cool periods as well, for even in the glacial periods the mixed feeder and browser dietary categories are present sporadically (**SZABÓ ET AL., 2021**).

VIII. It was concluded that the wear pattern of the warmer periods changes from a grazer signal to a mixed feeder and browser signal. This shift suggests an increase in the tree cover of the basin. The complexity of the available vegetation could have been significantly higher during the interglacials, because during these periods, almost all microwear dietary categories were observable. These conclusions agree well with results based on other vertebrate and mollusc faunas and palynological data (e.g. FŰKŐH ET AL., 1995; PAZONYI, 2011; MAGYARI ET AL., 2019) (**SZABÓ ET AL., 2021**).

IX. Based on the results of the microwear analysis conducted on 73 specimens, it was conlcuded that the different cervid species had markedly different dietary preference from each other. The elk and the reindeer specimens were characterized by a low-pit number mixed feeder diet, the irish elk with a mixed feeder diet, whereas the fallow and the roe deer were characterized with a browser diet. In contrast with the other species, red deer showed much larger dietary plasticity, because this species occupied a much larger area of the dietary morphospace (**SZABÓ ET AL., 2021**).

X. Based on the $\delta^{18}O_{PO4}$ values of the examined cervid teeth – originating from nine Pleistocene localities – average palaeotemperatures were calculated. These values seem to follow the global temperature trends. During the glacial periods, the reconstructed temperature was cooler than that of present day Hungary (up to 5-6 °C cooler during the LGM). Whereas, the interglacials were characterized by slightly warmer calculated temperature values (**SZABÓ ET AL., 2021**).

XI. Based on the $\delta^{13}C_{CO3}$ values of the examined enamel samples, it was concluded that the diet of the Quaternary cervids of the Carpathian Basin consisted mostly of plants utilising the C_3 photosynthetic pathway. The $\delta^{13}C_{CO3}$ values of the enamel samples ranged between -0.37‰ and -12.39‰. The highest value belonged to the reindeers from Szeleta-cave – possibly due to the increased consumption of lichens – whereas the lowest value belonged to an *M. giganteus* specimen from Tokod-Nagyberek I. (**SZABÓ ET AL., 2021**).

Publications related to the dissertation:

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