# Go the whole hog in microwear analysis! A new reference dataset of dental microwear textures in extant wild boars (Sus scrofa) and implications of intra-facet and intra-dentition variability for applications to the Mesolithic record

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**Résumé**

Zooarchaeological data available for Mesolithic sites from Quercy (France) fuel discussions on occupation periods and functional status of the sites within a logistical mobility system of those populations of hunter-gatherers. Dental microwear analysis of fauna has recently been proposed as an additional source of data on occupation time and seasonality of archaeological sites.

Three-dimensional Dental Microwear Texture Analysis (3D-DMTA) aims at quantifying microscopic wear features observed on enamel facets of occlusal surfaces of teeth. Microwear features result from attrition (tooth-to-tooth contacts), abrasion (tooth-to-food contacts, including exogenous particles sticking to food items), and erosion (from acidic foods). Microwear surfaces therefore reflect mechanical properties of ingested food items as well as mastication dynamics and dental morphology. Due to a rapid turnover, microwear surfaces observed on the teeth of a dead animal reflect a diet that is representative of the few days to few weeks before the death. As many animals display strong seasonal variations in diet, dental microwear analysis shows a high potential to reconstruct season of death, and therefore seasonality of human occupations at archaeological sites. A high microwear variation within a zooarchaeological sample would be expected for a long occupation or a repeated occupation at different times of the year whereas a low variation would be expected for samples resulting from short or seasonal occupations.

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Wild boars (*Sus scrofa*) are ideal targets for those microwear analyses as: 1) they are the dominant species in faunal spectra at most sites occupied by Mesolithic hunter-gatherers; and 2) their extant representants are characterized by opportunistic omnivorous diets with strong seasonal variations that reflect seasonal changes in vegetation. However, application of dental microwear analysis to Mesolithic wild boars is hindered by: 1) the lack of appropriate reference datasets on extant animals with precise temporal and geographic information; 2) the lack of knowledge on the various components (intra-facet, intra-tooth, intra-dentition) of variability of microwear signals; and 3) the fragmentary nature of the archaeological samples of Mesolithic sites, where most specimens are represented by isolated teeth representing different positions of the dentition. The objectives of our study are therefore: 1) to present a new reference dataset that could be applied to Mesolithic wild boars; 2) to better comprehend the impact of intra-facet and intra-dentition microwear variability on comparisons among specimens; and 3) to propose an optimal sampling strategy for future applications to archaeological contexts, especially to fragmented/isolated teeth that represent different positions along the dentition.

We took surface impressions of the occlusal surfaces of teeth using silicone elastomers. We measured surfaces of 333 x 351 $\mu$m on enamel shearing facets with the 100x objective of an optical profilometer and subjected the measured surfaces to several surface pretreatments. A 200 x 200 $\mu$m surface was then extracted, levelled, and subjected to Scale-Sensitive Fractal Analysis. To quantify differences in microwear textures, we compiled fractal parameters known to differ among mammals with different diets: complexity (Asfc), anisotropy (epLsar), heterogeneity of complexity (HAsfc-9 and HAsfc-81, respectively compiled using 9 and 81 subsquares), and textural fill volumes (Ctfv and Ftfv, respectively at coarse and fine scales). In nearly all previous studies, dental microwear parameters were compiled from a single surface (or a small number of surfaces, 2 to 4) per facet (hereafter called the "classic strategy"). This spatial localization of the surface within the facet is chosen almost randomly by the operator, with some consideration for possible preservation biases. This single surface is considered to be representative of the whole facet even though a facet is big enough to sample several dozens of surfaces and the degree of microwear variation within the facet is poorly known. We therefore explored for the first time intra-facet variability by conducting 3D-DMTA on 30 contiguous surfaces (six rows by five columns) that document around one third of the facet of a lower p4 of a Mesolithic wild boar from the Cuzoul de Gramat (Lot, France) Mesolithic site.

First, we quantified intra-facet variability by compiling coefficients of variation (CV) of the 30 surfaces for each microwear parameter. CV range from 28 % to 58 % (average = 47 %), indicating a strong intra-facet variation. Geary's C, an index of spatial autocorrelation, was compiled for each microwear parameter. Its values range from 0.87 to 1.12, suggesting that there is no spatial correlation of microwear parameters for that particular facet. If confirmed by additional specimens, it would indicate that operator-dependent randomly chosen spatial localization of the targeted surface within a facet will not significantly influence the comparisons among facets and specimens.

Secondly, we developed an original statistical approach to explore the impact of the observed intra-facet variation on comparisons among specimens when using a classic sampling strategy. How many 200 x 200 $\mu$m surfaces per facet would be sufficient to correctly estimate the facet mean of microwear parameters? For each sample size (ranging from 2 to 29), we randomly drew 5000 subsamples among the 30 surfaces. Mean value of each randomly chosen subsample was compared to the mean value compiled on the 30 surfaces of the facet by compiling the relative difference to the facet mean (RDFM), expressed as a percentage. As subsample size increases, the distributions of the 5000 values of RDFM get more symmetric and less dispersed. For each microwear parameter, we selected the RFDM thresholds that define the central 80 % of the RDFM values around the mode. Those interval widths decrease as the subsample size increases. For all microwear parameters, the amplitude is around 55 % for a subsample size of 5 surfaces (meaning that 80 % of the subsample means are at maximum 27.5 % higher or lower than the facet mean) and then quickly decreases to around 30 %, 25 %, and 20 % for subsample sizes of 10, 15, and 20, respectively. Depending on the expected differences between targeted specimens, the number of surfaces per facet required to obtain a precise estimate of the facet mean value will differ. A classic sampling strategy is likely confounding the detection of subtle microwear differences among samples. In future studies, it will be necessary to measure a larger number of surfaces per facet (> 10-15).

Thirdly, we explored the implications of that improved understanding of intra-facet variability when comparing two enamel facets using the classic sampling strategy. How much differences in microwear parameters would purely result from random sampling of a pair of surfaces within facets with strongly variable microwear textures? We considered two hypothetical facets whose microwear textures are strictly identical (by simply doubling the parameter values of the Cuzoul de Gramat lower p4 facet and considering it as two specimens). Among the 30 surfaces, we randomly sampled 500 pairs comprising one surface for each facet and compiled for each pair the absolute difference between the two values. For each microwear parameter, using the distribution of the 500 absolute differences, we identified the parameter threshold under which 80 % of the absolute differences are comprised and expressed it as a percentage of the whole range of intra-facet variation for that particular parameter. That percentage ranges from 33 % to 66 % (average = 48 %). Those high percentages indicate that, even when discarding the 10 % of highest and lowest absolute differences, numerous random pairs of surfaces would still detect relatively large differences between the two facets with identical microwear textures.
Finally, we explored intra-dentition variability by comparing microwear textures among different tooth positions in the lower dentition (p4, m1, m2) of a new reference dataset of wild boars, using the classic sampling strategy. We investigated a geographically and temporally well-constrained population of extant wild boars (*Sus scrofa*). Twenty-four individuals (including males and females, juveniles and adults) were collected by hunters in Tautavel (Pyrénées-Orientales, France), from August to November 2017. Based on the absolute differences between microwear parameters of m1 versus p4 and m1 versus m2, we explored their variation and compiled mean values of the absolute differences in tooth positions for the Tautavel sample. CV of intra-dentition differences range from 64 % to 117 % for m1-p4 and from 55 % to 113 % for m1-m2. Such high values of CV indicate a strong variability among individuals whereas mean absolute differences between tooth positions are relatively small. The percentages of the means of absolute differences relative to the whole range of values for each microwear parameter range from 11 % to 29 % for m1-p4 and from 6 % to 19 % for m1-m2. This suggests a relatively small difference among tooth positions and a slightly bigger difference between the premolar and molars than between the two molars. However, those intra-dentition differences appear relatively small compared to the differences that could be due to the random sampling of intra-facet variability, and therefore, based on the classic strategy, we cannot conclude on the reality of those intra-dentition differences. Implications for future studies of Mesolithic samples will be discussed.