A topographic approach to microwear analysis

Edward H. Hagen[1] and Phillip L. Walker[2]

[1]Institute of Theoretical Biology, Humboldt-Universität zu Berlin
Invalidenstraße 43, 10115 Berlin, Germany
e.hagen@biologie.hu-berlin.de

[2]Department of Anthropology, University of California, Santa Barbara 93106
pwalker@anth.ucsb.edu

Abstract

Studies of micron-scale surface wear patterns holds considerable promise as a source of functional information on both teeth and artefacts. Realizing this promise of microwear research has been hampered by fundamental limitations posed by the optical and scanning-electron micrographs used in virtually all microwear studies. The automated extraction of accurate 3D topographic information from single 2D photographs of surfaces is a formidable and largely unsolved problem. Consequently, microwear studies typically require that all microwear features be identified and measured by hand using time-consuming and often unreliable procedures. We review relatively new automated technologies for rapidly quantifying 3D surface topographies, and present a case study in which we replicate previous results using these new methods.

The Promise of Microwear Research

Studies of microscopic use-related surface modifications (microwear) on objects such as stone tools, ceramics, bones, and teeth have the potential to provide an enormous amount of valuable anthropological data. The microscopic modifications in surface topography considered in such studies range in spatial scale from sub-micron to approximately 1 mm. Although the techniques used in microwear studies vary, the vast majority of research has been conducted through the analysis of two-dimensional photographic images obtained using light and scanning electron microscopy.

Lithic microwear provides information on tool function and the behavior patterns associated with tool use. Studies of abrasions and cutmarks on bone provide evidence of early hominids subsistence activities, interpersonal violence, ritual processing of bodies and cannibalism.

Many researchers have studied dental microwear to obtain data on tooth function and diet (Gordon, 1982; Grine, 1981, 1986; Rensberger, 1978; Teaford, 1994; Walker et al., 1978). It has been shown that microwear can distinguish extant species with broadly different diets, such as frugivorous vs. folivorous anthropoids (Teaford & Walker, 1984) and grazing vs. browsing herbivores (Walker et al. 1978; Solounias & Moelleken, 1992; Solounias et al., 1988). Seasonal and ecological differences are also reflected in microwear patterns (Teaford & Glander, 1991; Ungar et al., 1995).

Dental microwear has also illuminated the diet of prehistoric and historic populations. Cereal and other plant-specific phytoliths have been shown to be important contributors to dental microwear and are associated with distinctive microwear patterns, for example, findings that point to the potential of dental microwear analysis to elucidate even relatively subtle aspects of diet (e.g., Danielson and Reinhard, 1998; Gügel et al. 2001). Dental microwear analysis has also been used to document age and sex-differences in diet (Pérez-Pérez et al., 1994; Pérez-Pérez et al., 1999), and has provided important information on dietary and non-dietary tooth use (e.g., Unger 1996 and references therein).

Despite these encouraging results, the development of microwear research in anthropology has been impeded by some fundamental problems associated with the quantification of differences in images of microworn surfaces. After reviewing problems, we will describe how they can be resolved using three-dimensional imaging techniques. We focus on dental microwear, but our conclusions apply to any study of the relationship between microwear and object use.

Limitations of Current Techniques

Missing information

Although early microwear studies were qualitative (Grine, 1981; Puech, 1979; Walker, 1981; Walker et al. 1978), it rapidly became apparent that most microwear patterns do not fall into discrete categories that can be correlated on a one-to-one basis with the processes that produced them. Patterns instead exhibit subtle, continuous variations that can only be resolved statistically (Gordon 1982, 1984; Grine 1986, 1987; Teaford 1985, 1986; Teaford and Walker 1984). Informative analyses of microwear therefore require accurate quantification.

Virtually all microwear studies use photographs (micrographs) of worn surfaces taken with cameras attached to optical or scanning electron microscopes (SEM). Photographs of microworn surfaces are a two dimensional representation of a three-dimensional surface topography.

Unfortunately, the topographical information contained in photographic images is inherently ambiguous. These images have a very complex, indirect, and inconsistent relationship to the topographic relief that is the subject of microwear analysis. Consequently, few, if any, quantitative studies of microwear have included any information on one of its key dimensions: feature depth. Because it is difficult to extract quantitative data on surface elevations from SEM images,[1] it is infeasible to study important aspects of microwear like peak height distributions or height-width ratios using them. Using current techniques, microwear analysts cannot quantify key aspects of how surfaces are altered by wear.

Subjectivity

Computer algorithms do not yet exist that can reliably distinguish artifacts like shadows from actual features like peaks and valleys. Microwear analysts therefore must rely on the subjective identification of features, which are then measured manually. This is both the strength and weakness of current techniques. It is a strength because the human visual system excels at identifying features in photographs. It is a weakness because subtle, often continuous, variations in microwear must be reduced to subjectively defined 'features' for measurement and quantitative comparison. This results in the loss of an enormous amount of information on subtle variations in topographic relief. A second problem arises from the fact that feature boundaries are frequently indistinct and overlapping; this opens the possibility for significant inter- and intraobserver measurement error. For example, the three most popular methods for quantifying dental microwear (Grine, 1986; Teaford, 1985; Unger 1995) all rely on subjective identification of features, which are then measured by assigning them four points representing the major and minor axes. The complex topography of a microworn surface is thus approximated as a set of overlapping rectangles of various lengths and widths. Two types of features are traditionally recognized: scratches, with a length to width ratio of > 4 to 1, and pits, with a length to width ratio of = 4 to 1. Using this approach, variations such as curved or irregular feature boundaries are ignored. This simplifies analyses, at the expense of ignoring potentially valuable topographical information.

Feature identification is time consuming

In dental microwear analysis, SEM micrographs are usually taken at a magnifications ranging from 100x to 500x. At the frequently used higher magnifications, a typical micrograph might have between 100 and 200 features that must be manually identified and measured (see, e.g., Grine et al. 2002). The time needed to identify and measure this many features obviously varies with the skill and experience of the microwear analyst; at best, though, it takes many minutes. The typical micrograph images an area of about 0.01-0.02 mm^2. Quantifying the microwear of a single wear facet of a single cusp of a single tooth would take hours. This opens the possibility for sampling biases because only a small area of a microworn surface is studied.

The time consuming nature of the current SEM based approach also severely limits the number of individual specimens that can be examined. Published dental microwear studies have so far been limited to a few individuals per species or population. Gordon's (1982) study of microwear in chimpanzee molars is a systematic attempt to assess the extent of variation within and among the dentitions of individuals from the same population. From her research, it is clear that functionally significant differences in microwear patterns are often present on the cusps of a

---

[1] We discuss an exception, stereoscopic SEM, below.

single tooth. Substantial differences also exist in the patterns present on different teeth in the same dentition, as well as between members of the same population. Studies by Teaford and Walker (1984) and Grine (1986) have confirmed the existence of significant intracrown and intrapopulation variations in microwear.

Impact of photographic variables

The visual identification of microwear features is influenced by all the variables involved in SEM imaging, including illumination, contrast, collector geometry, and magnification. Gordon (1988) has demonstrated significant differences in the statistics obtained from same microwear surface when photographed under different conditions. Magnification has a particularly profound effect on the number of features that an analyst will identify (but see Grine et al. 2002). “Image quality,” the result of several subjective decisions by the SEM operator, is another factor that is generally acknowledged as important.

Testing measurement reliability

Grine and co-workers (2002) conducted a study of inter-technique, inter-individual, and intra-individual error in dental microwear quantification that illustrates some of the strengths and weaknesses of current SEM based techniques. In this study, three popular and basically similar microwear quantification techniques (Grine, 1986; Teaford, 1985; Unger 1995) were used to assess the means of four microwear metrics (percentage of pits, mean pit length, mean pit breadth, and mean scratch breadth) on eight micrographs of different tooth surfaces. The analyst in each case was the inventor of the technique. The means for percentage of pits, a *key* microwear metric, were Grine M=45.3 (SD=10.0), Teaford M=27.9 (SD=10.1), and Unger M=53.7 (SD=10.8). Note that the mean value obtained by Unger is almost twice (1.92) the mean value obtained by Teaford, a highly significant difference (each used the same definition of a ‘pit’). On one micrograph, Unger and Teaford differed by a factor of 3.8 for this metric!

We reanalyzed the published inter-technique data using Pearson’s correlation coefficients, a simple and straightforward measure of reliability. Although the percentage of pits and scratch breadth measurements were moderately positively correlated for all analysts, none of the correlation coefficients were statistically significant, even if the level of significance was relaxed from .05 to .10. (Unger and Teaford were significantly correlated for scratch breadth, r=.55, p=.08. This correlation, however, was an artifact of a single outlier; with this outlier removed, r=.098, p=.42). Pit length and breadth were the only measurements for which there was significant agreement among all analysts (see table 1).

	Grine	**Teaford**	**Unger**
Percentage of pits			
Grine	1.0		
Teaford	.44 (p=.14)	1.0	
Unger	.37 (p=.18)	.40 (p=.17)	1.0
Mean scratch breadth			
Grine	1.0		
Teaford	.35 (p=.19)	1.0	
Unger	.39 (p=.17)	.55* (p=.080)	1.0
Mean pit length			
Grine	1.0		
Teaford	**.79** (p=.010)	1.0	
Unger	**.60** (p=.059)	**.85** (p=.004)	1.0
Mean pit breadth			
Grine	1.0		
Teaford	**.72** (p=.021)	1.0	
Unger	**.68** (p=.032)	**.89** (p=.001)	1.0

Table 1: Analyst correlation matrices for four microwear metrics measured on the same 8 micrographs; p's are one-tailed. Coefficients significant at the p<.10 level are in bold. The significant Unger-Teaford scratch breadth correlation (marked with an *) was an artifact of a single outlier. Computed from data in Grine et al. 2002.

Based on these results, Grine and co-workers rightly emphasize the need for standardized microwear quantification procedures. The inter-technique error of 19% that they found makes statistical comparisons of data produced using these slightly different procedures problematic. Inter- and intra-individual error results were more encouraging. When all analysts used Unger's 'semi-automated' technique involving his Microwear 4.0 software (and the same micrographs), no significant differences in these highly skilled analysts' mean metric values were found (the sample size of four micrographs, however, was quite small). Grine and co-workers estimate overall inter-individual error to be 9%, and intra-individual error to be 7%.

Previous automation efforts

Some work has been done to reduce these data collection problems by using automated computer image analysis systems (Walker et al. 1987, Ungar et al. 1991). Theoretically it should be possible to devise an automated system that rapidly does analyses that closely mimic those currently done by hand (e.g., identify, count and measure the microwear features present within the sampling field). The problem with this image analysis approach is that it is extremely sensitive to subtle variations in image quality. Even after extensive image processing to even out contrast and detect feature boundaries, two images of the same area of microwear taken on the same instrument under slightly different conditions can yield quantitative data that show statistically significant differences. With current techniques, microwear research has reached a "plateau" (Unger et al. 2003, p. 190).

Advantages of the Topographic Approach

There is a consensus among microwear researches that solutions to the problem of laboriously digitizing subjectively identified microwear features need to be found if microwear analysis is to reach its full potential (Teaford 1994; Walker and Teaford 1989). The topographic approach we discuss here promises to help resolve these difficulties. Instead of attempting to extrapolate three-dimensional information from two-dimensional images, we and other microwear researchers advocate the use of high-resolution surface mapping technologies to directly study the three-dimensional topography of a surface (Walker and Hagen 1994; see also Dennis et al. 2004; Hillson and Jones 1989; Ungar and Williamson 2000; Unger et al. 2003). In engineering, the study of surface topography is termed *surface metrology* (e.g., Whitehouse 1994). Many different techniques have become available in the last several years that allow surface topography to be rapidly quantified and characterized. Some of the most promising of these are discussed below.

Within the resolution limits of the instrument used to produce them, the surface maps produced by these new techniques provide comprehensive, unambiguous descriptions of an object's surface topography. Because these topographical maps are a direct representation of vertical relief (z) as a function of x and y coordinates, they allow the object's surface to be characterized statistically without the subjective and time-consuming step of manual feature identification. Surface statistics computed from them provide summary information on features at all scales. This completely eliminates the quantification problems arising from the many variables that alter the appearance of optical and SEM micrographs. Because complete information has been collected and subjective feature identification has been eliminated, different researchers can easily and reliably compare their results.

The topographic approach is also often (though not always) significantly faster than the current SEM based approach. With some technologies, such as optical interferometry, topographies of areas roughly equivalent to those used in SEM microwear studies can be mapped and surface statistics computed in a minute or two. This allows much larger sample sizes to be analyzed than has been possible in the past. With other technologies, scanning times are longer, but still less than the semi-automated SEM quantification techniques; all non-SEM technologies also eliminate the extra time needed to prepare SEM samples and take SEM micrographs.

High resolution surface mapping instruments are in wide use in material science research and manufacturing, and a large number of devices are available 'off-the-shelf' complete with sophisticated software packages designed to analyze and characterize surface topography. We have investigated a variety of surface mapping technologies to determine their suitability for microwear research. There are two basic types: those based on a scanning stylus, and those based on optical reflection or transmission. The stylus-based technologies include atomic force microscopy and stylus profilometry, and the optical technologies include optical interferometry, confocal microscopy, and microcomputer tomography; stereoscopic SEM is a third type. Each technology has potential applications in microwear research, and we give a brief overview of them here.

Instrument	**Vertical resolution**	**Vertical range**	**Lateral resolution**	**Lateral range**	**Slope limit**	**Notes**
Atomic force microscope	0.05nm	5-10µm	0.001µm	100µm	>70	Vertical range unsuitable for all but the highest magnification studies of tooth surfaces. Widely used in surface studies.
Stylus profilometer	1nm (0.1nm in some models)	130µm (up to 1000µm in some models)	0.05µm	>25000µm	45	Stylus pressures unsuitable for polymer casts and other delicate materials. Relatively inexpensive. Widely used for surface metrology.
Vertical scanning interference microscope	0.05nm	100µm (up to 5000µm in some models)	0.3µm minimum (limited by wavelength of light)	Depends on power of objective lens; at high power: 160µm	36	Slope limit and high surface reflectivity requirement prevent use with very rough and/or unreflective surfaces. Widely used for surface metrology.
Confocal scanning microscope (reflection)	500nm (5nm in models specialized for measuring surface heights)	>1000µm	0.2µm	Depends on power of objective lens; at high power: 160µm	high	Some versions of the technology are relatively inexpensive (~$50K), but the versions most suitable for dental microwear research are expensive (~$2-300K). Increasingly used for surface metrology.
Micro CT	5000nm	>>1000µm	5µm	>>1000µm	100	Maximum resolution is currently too low for detailed microwear studies. Cost is high (~$200K). Not currently used for surface metrology, to our knowledge.
Stereoscopic SEM	1nm	>>1000µm	.001µm	>>1000µm	100	Increasingly used for surface metrology.
Ideal dental microwear microscope	*1-10nm*	*>200µm*	*0.001-0.010µm*	*1000µm*	*>70*	

Table 2: Typical characteristics of different instruments for measuring surface topographies

Atomic force microscopy

Atomic Force Microscopy (AFM) is one of a family of methods, collectively called Scanning Probe Microscopy (SPM), for imaging surfaces with atomic or near-atomic resolution (related methods are Scanning Tunneling Microscopy, Lateral Force Microscopy, and Magnetic Force Microscopy). SPMs, including AFMs, have the highest spatial resolution of the currently available devices. These instruments have been used, for example, to directly measure the three-dimensional structure of DNA molecules. We found that the data we obtained using an AFM is of limited value for addressing the kinds of problems that have traditionally concerned anthropologists. Most worn artifact and tooth surfaces, for example, are far rougher than those AFMs are designed to analyze. AFMs generally can image surface features with heights no greater than 5-10µm, yet a single large microwear scratch may be this deep. Given that the tooth surface itself can slope tens of microns over the scanned area, AFMs are a less-than-ideal choice for most dental and lithic microwear studies.

In spite of these problems, we believe that AFM data may prove useful for some types of microwear research. Based on our stylus profilometer and interference microscope studies we have identified asymmetries and other shape irregularities in microwear pits and scratches that appear to contain information on direction of movement as well as other wear process variables. Extremely high resolution AFM data on the edges and sides of microwear features are likely to be useful for exploring these important aspects of microwear production.

Stylus profilometry

In stylus profilometry, a diamond stylus is dragged across a surface and the signal generated by its rise and fall is used to describe the topography of a surface transect. By scanning a series of such transects, this technique can be used to generate three-dimensional surface models. A significant advantage of stylus profilometers is their horizontal range, often on the order of centimeters, and their vertical range, which can be as high as 1mm.

The main disadvantage of stylus profilometry in microwear research is the large force exerted on the surface of the specimen by the stylus. In our experience, when properly adjusted, these instruments work well on enamel, obsidian, and other hard surfaces. When used to scan epoxy surface replicas, in contrast, there is an attenuation of the profilometer signal because the stylus scratches the replica’s surface. This is a significant limitation for microwear research because it is often not feasible to examine specimens directly. Scan times for this technology are significantly longer than for other technologies.

Optical interferometry

Interference microscopes exploit the fact that when two light waves are brought together they interact (Robinson et al. 1991). If they are in phase, they reinforce each other, if they are out of phase they cancel each other out. In such a microscope, a beam of light is split. One of the resulting beams is reflected off the specimen and the other off a reference mirror. When the two beams are recombined, the amount that they are out of phase will be proportional to the difference in the distances they have traveled. This difference, in turn, is determined by variation in the topographical relief of the specimen. With the aid of modern video chips and microcomputers, it is possible to record differences in light intensity associated with the phase shifts and convert them into an extremely accurate map of the surface’s microtopography (see table 2).

The variation of this technique most suited to surfaces with a significant amount of vertical relief (e.g., dental microwear) is called *vertical scanning interferometry*. In these instruments, the microscope head scans vertically while the interference image is focused onto a 256x256 element or similarly sized photo-detector. Computer algorithms then transform the three-dimensional interferogram into a quantitative 3D image. The process takes less than a minute.

The principle disadvantage of these instruments is that they require that sufficient light to form an interference pattern be reflected back through the lens system by the specimen *at each point.* If, at any point on the specimen within the field of view, the specimen is highly sloped, light will be reflected away from the vertical, and therefore will not be transmitted back through the lens system. At these points, such as the steep walls of a microwear scratch, the instrument will be unable to provide any information about surface height. Maximum imagable slope angles are less than 40 degrees. The same problem occurs if the object being imaged has low reflectivity at any point. In our experience, as much as 5-10% of a typical microworn tooth surface may fail to image properly (special specimen coatings, however, might reduce the problem).

Confocal microscopy

In a conventional microscope, light from regions of the specimen in the focal plane, as well as light from regions above and below the focal plane, reach the eye. Regions in the focal plane are in-focus, and those above and below the plane are out-of-focus. By inserting a diaphragm with a pinhole between the lens and the eye (or other photodetector), it is possible to admit only rays of light from the point of the specimen at the focal point (in the center of the focal plane), thereby excluding all light from out-of-focus regions. Then, by moving the specimen on the stage, or by illuminating the specimen through the microscope optics with a scanning light beam (e.g., a laser reflected off oscillating mirrors), and recording the intensity of light passing through the pinhole, it is possible to generate an image of only those regions of the specimen in the focal plane, i.e., those regions that are in-focus. Finally, translating the specimen vertically in small increments and repeating the image-scanning process at each step can capture a detailed 3D image of the specimen. If the specimen is at all transparent (like living cells, where this technique is often used), the data collected will represent the volume distribution of the specimen; otherwise it will represent the surface topography. The versions of these microscopes most suitable for dental microwear research can be quite expensive (~$2-300K).

Micro computed X-ray tomography

The familiar X-ray computed tomography (CT) scanner has recently been miniaturized to desktop size and the resolution has been increased tremendously from voxels (the cubic equivalent of pixels) in medical devices that are approximately $1mm^3$ in size to voxels as small as 5 microns on a side. In these micro CT scanners with such high resolutions, a thin, horizontal fan of x-rays is projected through a turntable-mounted specimen to an array of detectors on the other side. After a complete rotation of the specimen, a high-resolution map of the cross-section of the specimen can be reconstructed from the pattern of x-ray intensities sampled at the detectors. By translating the turntable vertically in small increments and repeating the rotation and scanning process, a stack of cross-section ‘slices’ is produced which provides a complete 3D map of the both the surface and internals of the specimen. Computer software can then section the specimen through any plane and rotate it so that its surface can be viewed from any perspective.

With current technology, the internal and external structures of a small object can be completely scanned at near-micron resolutions. Unfortunately, some of the finest microwear scratches of anthropological interest are about 1 micron wide by 1 micron deep, so this technology is only appropriate for more coarse-grained studies, such as macro wear patterns, as well as volumetric studies of wear regions. Micro-CT resolution is currently most limited by the spot size of the x-ray source (currently about 5µm). If and when x-ray sources with sub-micron spot sizes are produced, micro-CT might become a viable technology for microwear research. (To our knowledge, micro-CT has not currently been used for surface metrology, and will probably remain best suited for studies of the three-dimensional distributions rather than surface structures.)

Stereoscopic SEM

The high resolution, high depth-of-field images produced by SEM are ideally suited for stereophotogrammetric analyses. Digital elevation models of imaged surfaces can be produced using a stereo-pair of ordinary SEM images (Boyde 1973; Howell and Boyde 1972; Minnich et al. 1999). A specimen is photographed using standard SEM procedures; the specimen is rotated a few degrees and photographed again. A large number of homologous points on the pair of images are identified using sophisticated image-analysis algorithms, and a digital elevation model is then easily computed if the angle of rotation and a few other simple parameters are known. The resolution of this model is the same as the resolution of the SEM images.

Although the difficulty of identifying features in an SEM image using a computer algorithm is exactly the problem that motivated us to investigate other imaging technologies, the problem of identifying homologous features on a very similar pair of images that differ from each other only by a simple rotation is considerably simpler than the general problem. This step is still quite computationally expensive, however, and can take from minutes to hours depending on the size of the images and the speed of the computer.

An advantage of this method is that it only requires access to a standard SEM, which can be found on almost every university campus, an appropriate software package, and a desktop computer. In practice, the SEM must possess a high-quality stage so that the specimen can be rotated accurately and eucentrically (i.e., the midpoint of the specimen surface being imaged must be positioned so that it is centered on the axis of rotation); it should also be able to output digital image files.

Surface characterization

One of the principle advantages of topographic approaches is that once surface topography has been quantified, most standard statistical procedures can be used to analyze the data, including basic statistics such as the standard deviation of the surface’s height (see table 3 for a partial list). It is not clear which, if any, of the methods listed in table 3 will prove useful for microwear analysis. We anticipate that several will be necessary to characterize microworn surfaces in ways that illuminate diet and tooth use. Almost none of them, we note, can be used with the current single-image SEM procedures.

Statistic	**Use**	**Reference**
Residual surface ?(x,y): The resulting data set after filtering, if any, has been applied. Filtering may include tilt, curvature, etc.	Most tooth surfaces will have large-scale features like curvature from crown morphology that are not of direct interest for microwear studies. These must often be filtered out prior to analyses. Sometimes this is done by subtracting a first or second degree polynomial from the data; other times a high-pass filter is employed.	This, and the following S-parameters, were defined in 1991 by the attendees of the first EC Workshop on 3D Surface Measurement and Characterization.
Roughness average: S_a (termed R_a when the surface is not filtered). The arithmetic mean of the absolute values of deviations from the mean plane of the residual surface.	A measure of the roughness of the residual surface. Units are usually in nanometers or microns.	Ibid
Root mean square (RMS) roughness: S_q (termed R_q when the surface is not filtered). The square-root of the mean squared values of the residual surface height data.	The standard deviation of the height data from the residual surface. Units are usually in nanometers or microns.	Ibid
Skewness: S_{sk} (termed R_{sk} when the surface is not filtered). A measure of asymmetry of the residual surface about the mean plane; alternatively, the asymmetry of the height distribution.	Smooth surfaces with bumps have positive skewness, whereas smooth surfaces with scratches and pits have negative skewness.	Ibid
Kurtosis: S_{ku} (termed R_{ku} when the surface is not filtered). A measure of the 'shape' of the height distribution (i.e., broad with short tails, or spiky with long tails).	Height distributions with long tails (i.e., many tall peaks and deep valleys) will have high kurtosis, whereas those with short tails will have low kurtosis. Kurtosis is also a measure of deviation from a normal distribution. It is sensitive to outliers; others measures of non-guassianity like negentropy are more robust.	Ibid
Correlation function	Degree of self-similarity of a surface.	Church and Takacs 1988
Height histograms; peak-valley distributions and ratios	Can reveal clustering of feature sizes and other straightforward aspects of surface topography. Distributions can be characterized using standard descriptive statistics.	
Roughness exponent	It has been hypothesized that the relationship between the width and length of fractured surfaces goes as $w \sim L^x$, where x is a universal roughness exponent, independent	Hansen et al 1991

	of material.	
Structural anisotropy	Microwear may be “oriented” in a particular direction. Statistics computed along transects taken in one direction will then differ from those computed along transects taken in other directions.	Kanatani 1984
Fourier Amplitude and Power spectrums	A complex waveform can be represented as a sum of sines and cosines of different frequencies and amplitudes. The Fourier spectrum is simply the distribution of amplitudes of the different frequency components. For microwear, the frequencies are spatial frequencies, so the Fourier spectrum is—roughly—the distribution of feature sizes.	Church and Takacs 1988
Amplitude:wavelength ratios	Gives the rms roughness within a spatial frequency octave.	Power and Tullis 1991
Fractal dimension	How rough a surface appears when measured at different scales.	Power and Tullis 1991
Crossover dimension	When a profile is measured at scales greater than the crossover dimension, it appears flat, whereas when measured at scales less than the crossover dimension, it appears rough.	Power and Tullis 1991
Total profile variance	Steepness of the topography	Power and Tullis 1991
Scale-area relations **Scale-length relations**	Relationship of surface area to the scale at which it is measured. Particulars of this relationship can yield clues to surface formation processes.	Brown, et al 1993
Principle Components Analysis (PCA)	A standard method for reducing a large number of variables to a smaller number of uncorrelated factors. Often used in image analysis.	Jackson 1991
Independent Component Analysis (ICA)	A standard method for decomposing a complex signal that is the sum of two or more statistically independent signals into its independent components. Might be able to decompose a microwear pattern into wear patterns caused by separate wear processes.	Hyvärinen et al. 2001
Wavelets	Like Fourier analysis, wavelet transforms can extract salient features from a complex signal. Unlike the sines and cosines of Fourier analysis, which vary in frequency but are infinite in extent, wavelet functions vary in both	Burrus et al. 2001

	frequency and in spatial scale, so they are better able to represent waveforms with sharp spikes and discontinuities.	

Table 3: Some quantitative approaches to characterizing surface topography.

Pilot study

To determine whether a relatively inexpensive, off-the-self surface profiling instrument (a stylus profilometer) could provide information useful for microwear analysis, we conducted a pilot study using uniform, homogeneous surfaces abraded under controlled conditions. Our study was, in part, a replication of one aspect of a previous controlled *in vitro* tooth abrasion study by Maas (1994). Using the standard procedure of measuring features subjectively identified through the visual inspection of SEM micrographs, Maas found that under compressive loads, abrasive particle size was the principle determinant of pit size. Our goal was to replicate her results using a procedure that eliminated the subjective step of visually identifying microwear features. We did this through the statistical analysis of quantitative data on the topography of experimentally abraded surfaces.

Materials and methods

Unlike Maas (1994), we chose to abrade standard borosilicate glass microscope slides instead of teeth in order to reduce the number of variables we would have to control for. Specifically, we first wanted to ensure that the surfaces being abraded were initially identical. Second, we were interested in analyzing the microworn surface without the additional complication of compensating for gross changes in crown surface topography. Third, dental enamel is composed of calcium hydroxyapatite crystals about 50nm in diameter that are bundled in prisms several microns in diameter. The latter are detectable at the sub-micron resolutions we intended to explore, whereas glass is structurally homogeneous at these resolutions. Previous studies had also found that, under shear loads, wear features were influenced by the orientation of enamel crystallites (see Mass 1994). By using glass slides, we avoided the additional complications that enamel microstructure might impose on our analyses.

We abraded the glass slides in two abrasive conditions—free and fixed abrasive—using three different grit sizes in each condition. To ensure a relatively uniform abrasion process, we constructed a reciprocating mechanism that moved the glass slide a fixed distance (5 cm) laterally on an abrasive surface with 5 kilograms of vertical (compressive) force. Glycerine was used as a lubricant for both fixed and free abrasives because of its relatively high viscosity and water solubility (which facilitated cleanup).

In the free-abrasive condition, silicone carbide powder in one of three grit sizes (220, 400, 600)[2] was mixed with glycerine to form a slurry. The slurry was sandwiched between two glass slides, and the top slide was abraded against the bottom slide. In this condition, the load of the grit on the slide is almost entirely compressive. Fresh abrasive was used for each slide. In the fixed-abrasive condition (not studied in Maas 1994), the abrasive surface consisted of silicon carbide wet-or-dry sandpaper in the same three grit sizes: 220, 400, and 600. In this condition, there are both compressive and shear loads between the grit and slide (Maas 1991) Fresh sandpaper was used for each slide. All slides were 'stroked' 20 times in the reciprocating device,

---

[2] Although we did not measure the distribution of particle sizes of these grits ourselves, published size ranges are similar to the grit sizes used by Maas. See table 5.

and then rinsed and labeled. We abraded seven slides in each of the six abrasive conditions for a total of 42 slides.

Slides were visually inspected using an optical microscope at low and medium powers, and then scanned using a Dektak 3ST stylus profilometer at its highest lateral resolution, 0.25µm per data point. This resolution is comparable to the resolution used in typical SEM microwear studies. The radius of our stylus tip was 2.5 microns, so we were in effect collecting a moving average over a 2.5 micron region (tips with a radius of 0.08 microns are available for this model). The vertical resolution is claimed to be less than 1 nanometer (the maximum vertical surface variation tolerated by this instrument is 100 microns, which effectively limits scan lengths on curved surfaces such as teeth). Unlike some stylus profilometers, the Dektak 3ST was not designed to scan areas, only single, linear transects of a surface. The sample is placed on a rotating, x-y stage, and is viewed using a video microscope in order to identify the region of interest. A single two millimeter scan of the middle of each slide was taken perpendicular to the direction of abrasion, for a total of 8000 points per scan. No sample preparation was necessary, and instrument setup time was minimal. Each scan took about 1 minute, and was de-trended using the instrument data acquisition software. Several unabraded slides were scanned to determine initial surface roughness. No vertical surface deviations larger than 10 nanometers were detected. Data were processed using Matlab, DADiSP, and SPSS.

Results

Visual inspection

Visual inspection of the abraded slides using a lower power optical microscope reveals a dramatic difference between slides abraded with fixed abrasive versus those abraded with free abrasive. Surface features of slides abraded with fixed abrasive consisted almost entirely of long, linear scratches, whereas surface features of slides abraded with free abrasive consisted almost entirely of roughly circular or elliptical pits. This effect has been previously reported by Lawn and Wilshaw (1975:1076); Maas (1994) similarly found that the microwear features of teeth abraded under compressive loads consisted largely of pits.

Stylus profilometry

Surface profile data were imported into DADiSP, a spreadsheet-style digital signal processing program that can display and analyze waveform data (see figure 1).

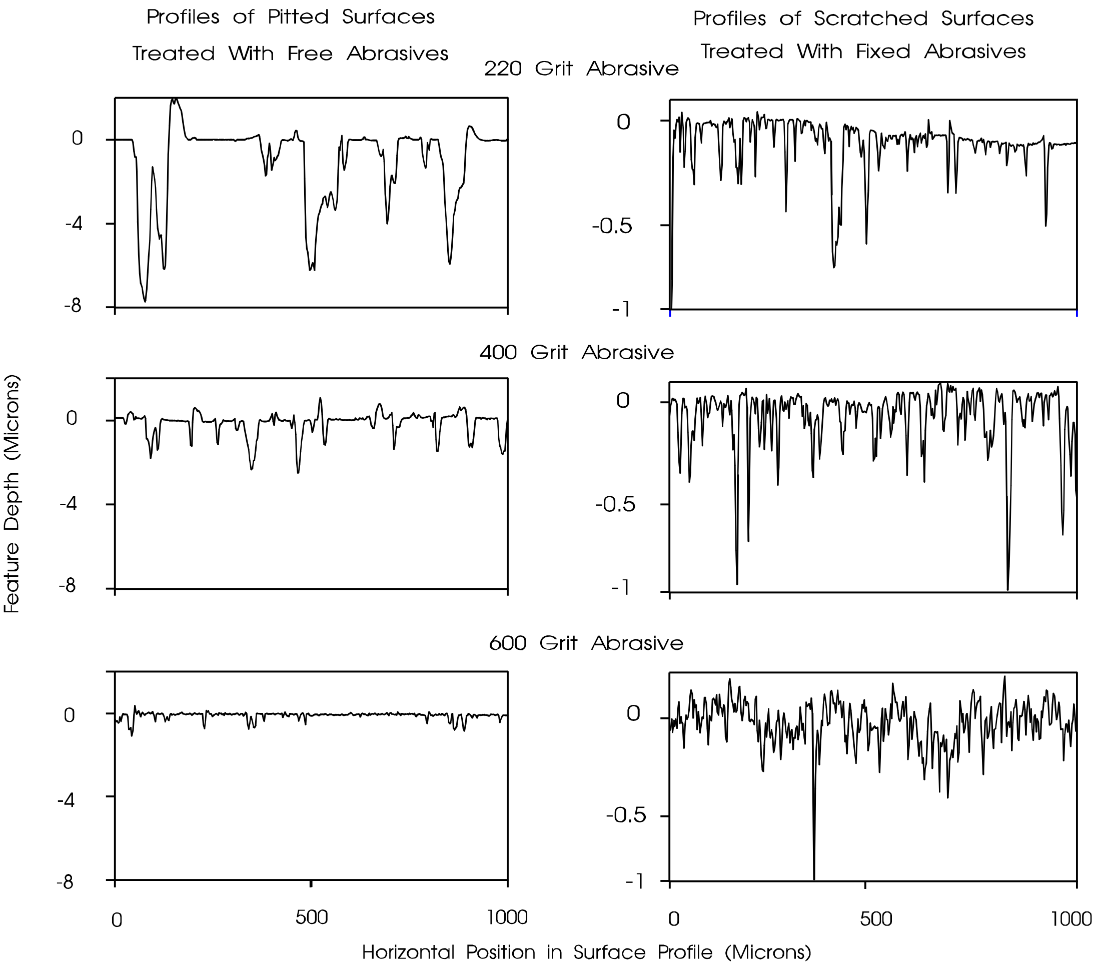

Figure 1: Typical surface profile of a glass slides abraded with free abrasive (left hand side) and fixed abrasive (right hand side). The microwear features of slides abraded with free abrasive consisted almost entirely of pits, whereas those abraded with fixed abrasive consisted almost entirely of scratches.

We elected to analyze the surface data using Fourier spectral analysis, computing the Fourier spectrum of each surface profile after first applying a standard Hamming window[3] (see figure 2). These spectra are frequency distributions for each surface profile, in which low frequencies represent large features and high frequencies small features. They can therefore be interpreted as distributions of feature sizes, and standard descriptive statistics of the distributions like mean, variance, and skew can be computed.

---

[3] A Hamming window tapers the data at the beginning and end of the waveform to zero in order to prevent the length of the waveform itself being interpreted as a component of the spectrum.

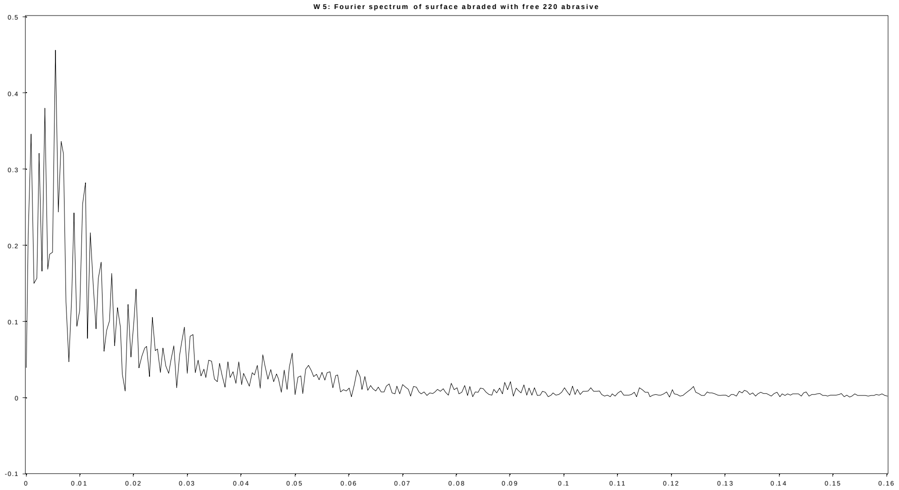

Figure 2: Fourier spectrum of the 220 pitted surface data depicted in figure 1. This spectrum is a distribution of the sizes of microwear features. The peak in the spectrum on the left-hand side corresponds to a feature size of 182µm.

Mean feature sizes

We computed the mean spatial frequency <w> from the Fourier spectra P(w) of each of the 42 abraded slides using the following formula:

$$\langle w \rangle = \frac{\int wP(w)\, dw}{\int P(w)\, dw}$$

The mean spatial period, or feature size, is simply <t>=1/<w>. Then we produced 'box-and-whisker' plots of the means of each of the seven slides within an abrasive condition (e.g., all slides abraded with free 220 abrasive). The results are depicted in figure 3; grit size distributions are in table 4.

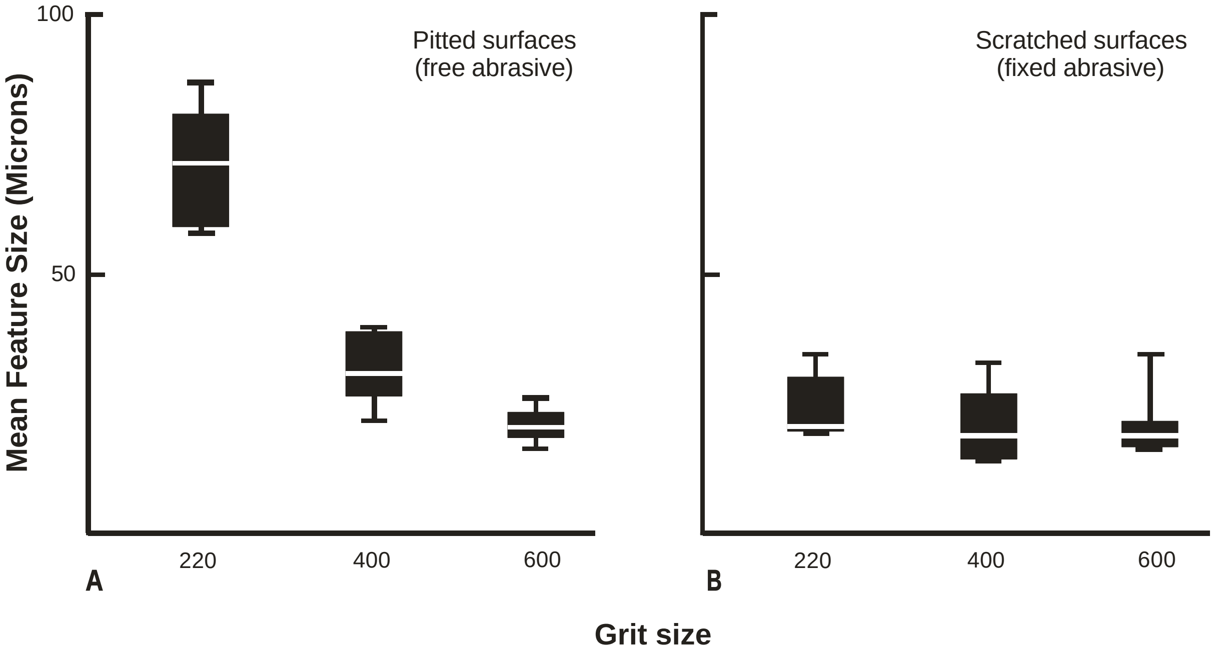

Figure 3: Box-and-whisker plots of the distribution of mean feature sizes for all seven slides abraded with a given grit size of free abrasive (A) and fixed abrasive (B).

As can easily be seen in figure 3A, there is a strong relationship between mean feature size and grit size when surfaces are abraded with free abrasive (compressive load), as Maas found. By the Mann-Whitney U-test, the mean pit sizes for 220 grit are significantly larger than those for 400 grit ($p<.001$), and those for 400 grit are significantly larger than those for 600 grit ($p<.01$). Figure 3A, then, is a replication of Maas' 1994 result that did not depend on subjectively identifying microwear features. This result does not hold for surfaces abraded with fixed abrasive (shear and compressive loads): there were no significant differences for the distributions of mean features sizes for the scratched surfaces (fig. 3B).

Grit size	**Approximate range of particle sizes**	**Range of mean feature sizes (free)**	**Range of mean feature sizes (fixed)**
220	60-75	58-87 (median=72)	18-34 (median=20)
400	20-23	22-40 (median=31)	16-33 (median=19)
600	13-16	17-26 (median=20)	15-35 (median=19)

Table 4: Range of grit particle sizes and mean feature sizes of abraded surfaces in microns.

Discussion & conclusion

Maas (1994) found that, under compressive loads, prepared dental enamel specimens abraded with different sizes of silicon carbide grits formed pits whose sizes were primarily determined by grit size. Microwear features (e.g., pits) were identified subjectively. We have replicated this result with glass surfaces using a method that did not require the subjective identification of microwear features. We also found that this result does not hold for glass surfaces abraded with a fixed abrasive—in this case, feature sizes were independent of grit size. A possible explanation is that, under shear loads, wear is principally caused by a single point contact between a silicon carbide grit particle and the glass substrate. In these circumstances, the size of the abrasive particle is irrelevant since only the point of the particle is causing the wear.

The applicability of our results to dental microwear studies is limited by our use of glass instead of dental enamel, and by our use of an extremely hard abrasive—silicon carbide—that is unlikely to be found in natural diets.

We argue, more generally, that microwear research will benefit from the use of surface mapping technologies that are able to provide quantitative, high-resolution information on surface height, information that does not depend on microscope settings or on subjective identification of features. The availability of such information opens the door to the use of a large and growing number of statistical and signal processing techniques for detecting, analyzing and classifying not only dental microwear features, but also lithic microwear features and surface features of any material that can be imaged with surface mapping instruments.

Our discussion of surface mapping technologies omitted the many difficult issues of measurement error, linearity, repeatability, imaging artifacts, etc., that must be addressed before any such technology can be used with confidence. Although we firmly believe the benefits of using these technologies will outweigh the costs, it would be unwise to underestimate the effort required to lay the necessary groundwork. In this regard, much work has already been done to assess the strengths and weakness of most of the technologies we discuss. Dental and lithic microwear research would do well to forge stronger connections with the burgeoning fields of surface metrology (e.g., Whitehouse 1994) and tribology (in part, the study of wear; e.g., Hutchings 1992; Rabinowicz 1995), fields that have done much to develop and refine surface measurement and have also made significant advances in the characterization of surfaces and wear processes.

Because most surface mapping instruments are both expensive and less common in university laboratories than are SEM’s, we suggest that, in the short term, microwear analysts investigate stereoscopic SEM. Although at one time this technique was time consuming and expensive (Gordon 1988), recent increases in computer power and image processing capabilities make it attractive. It requires only a few extra minutes to take an additional SEM image that is rotated with respect to the first image. Commercial packages that run on standard desktop computers are available to construct digital elevation maps from the stereo image pair (one such package, MeX, which we have not evaluated, is available from Alicona Imaging; a demo version can be downloaded at http://www.alicona.com/).

Finally, we advocate a shift in emphasis from the ‘two box’ strategy that is frequently pursued in dental microwear research (figure 4a), to a ‘four box’ strategy (4b).

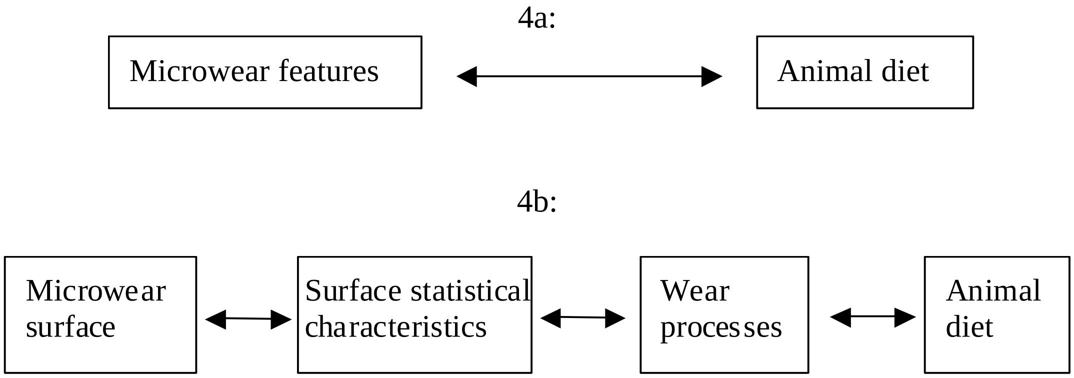

Figure 4: Two dental microwear research strategies

Much useful work has been done following the ‘two box’ strategy, but the work that has been done following the ‘four box’ strategy (e.g., Maas 1991, 1994) indicates that the ‘two box’ strategy might have fundamental limits. It may be almost impossible to link microwear features with animal diets, at least in some cases, without first developing both a richer suite of statistical surface characteristics, and a better understanding of the relationship between these characteristics and different wear processes caused by distinct aspects of an animal’s diet.

Acknowledgements

We gratefully acknowledge the generous donation of instrument time by Veeco Instruments, Santa Barbara, CA.

Literature cited

Boyde, A., 1973, Quantitative photogrammetric and qualitative stereoscopic analysis of SEM images, *J. Microscopy*, 98, 452-471.

Brown, C. A., Charles, P. D., Johnsen, W.A., and Chesters, S., 1993, Fractal analysis of topographic data by the patchwork method, *Wear,* 161, 61-67.

Burrus, C. S., Gopinath, R. A., Guo, H., 2001, *Introduction to wavelets and wavelet transforms: a primer*. N.J.: Prentice Hall.

Church, E. L., Takacs, P. Z., 1988, Instrumental effects in surface finish measurements, *Surface Measurement and Characterization,* 1009, 46-55.

Danielson, D. R. and Reinhard , K. J., 1998, Human Dental Microwear Caused by Calcium Oxalate Phytoliths in Prehistoric Diet of the Lower Pecos Region, Texas, *American Journal of Physical Anthropology,* 107, 297–304.

Dennis, J. C., Ungar, P.S., Teaford, M.F. and Glander, K. E., 2004, Dental topography and molar wear in Alouatta palliata from Costa Rica, *American Journal of Physical Anthropology*, 125, 152-161.

Gordon, K. D., I982, A study of microwear on chimpanzee molars: implications for dental microwear analysis, *American Journal of Physical Anthropology*, 59, 195-215.

Gordon, K. D., 1984, Hominoid dental microwear: Complications in the use of microwear analysis to detect diet, *J Dent Res,* 63, 1043–1046.

Gordon, K. D., 1988, A review of methodology and quantification in dental microwear analysis, *Scanning Microsc*, 2, 1139-1147.

Grine, F. E., 1981, Trophic differences between 'gracile' and 'robust' australopithecines: a scanning electron microscope analysis of occlusal events, *South African Journal of Science,* 77, 203-230.

Grine, F. E., 1986, Dental evidence for dietary differences in *Australopithecus* and *Paranthropus*: a quantitative analysis of permanent molar microwear, *Journal of Human Evolution,* 15,783-822.

Grine, F. E., 1987, Quantitative analysis of occlusal microwear in *Australopithecus* and *Paranthropus*, *Scan Microsc*, 1, 647–656.

Grine, F. E., Ungar, P. S. and Teaford, M. F., 2002, Error Rates in Dental Microwear Quantification Using Scanning Electron Microscopy, *Scanning,* 24, 144–153.

Gügel, I. L., Gruppe, G. and Kunzelmann, K. H., 2001, Simulation of Dental Microwear: Characteristic Traces by Opal Phytoliths Give Clues to Ancient Human Dietary Behavior, *American Journal of Physical Anthropology*, 114, 124 –138.

Hansen, A., Hinrichsen, E.L., Roux, S., 1991, Roughness of Crack Interfaces, *Physical Review Letters,* 66, 2476-2479.

Hillson, S. W. and Jones, B. K., 1989, Instruments for measuring surface profiles: An application in the study of ancient human tooth crown surfaces, *Journal of Archaeological Science*, 16, 95-106.

Howell, P.G.T. and Boyde, A., 1972, Comparison of various methods for reducing measurements from stereo pair scanning electron micrographs to "real data", *Scanning Electron Microscopy,* IITRI (ed. by 0. Johari), pp. 233-240, IIT Research Chicago.

Hutchings, I. M., 1992, *Tribology: Friction and wear of engineering materials*. Boca Raton: CRC Press.

Hyvärinen, A., Karhunen, J. and Oja, E., 2001, *Independent Component Analysis*. John Wiley & Sons.

Jackson, J. E., 1991, *A user's guide to principal components*. New York: Wiley.

Kanatani, K., 1984, Stereological Determination of Structural Anisotropy, *International Journal of Engineering Science,* 22, 531-546.

Lawn, B., Wishaw, T. R., 1975, Indentation fracture, *J Mat Sci*, 10, 1049-1081.

Maas, M. C., 1991, Enamel structure and microwear: an experimental study of the response of enamel to shearing forces, *American Journal of Physical Anthropology*, 85, 31-49.

Maas, M.C., 1994, A scanning electron-microscopic study of in vitro abrasion of mammalian tooth enamel under compressive loads, *Archives of Oral Biology,* 39, 1-11.

Minnich, B., Leebj, H., Bernroider, E.W/N. and Lametschwandtners, A., 1999, Three-dimensional morphometry in scanning electron microscopy: a technique for accurate dimensional and angular measurements of microstructures using stereopaired digitized images and digital image analysis, *Journal of Microscopy,* 195, 23-33.

Pérez-Pérez, A., Lalueza, C., Turbon, D., 1994, Intraindividual and intragroup variability of buccal tooth striation pattern, *American Journal of Physical Anthropology,* 94,175-187.

Pérez-Pérez, A., Bermúdez De Castro, J.M. and Arsuaga, J. L., 1999, Nonocclusal Dental Microwear Analysis of 300,000-Year-Old *Homo heilderbergensis* Teeth From Sima de los Huesos (Sierra de Atapuerca, Spain), *American Journal of Physical Anthropology,*108, 433–457.

Power, W.L., Tullis, T. E., 1991, Euclidean and Fractal Models for the Description of Rock Surface Roughness, *Journal of Geophysical Research,* 96 (B1), 415-424.

Puech, P. F., 1979, The diet of early man: Evidence from abrasion of teeth and tools, *Current Anthropology,* 20, 590–592.

Rabinowicz, E., 1995, *Friction and wear of material*s. New York: Wiley.

Rensberger, J. M., 1978, Scanning electron microscopy of wear and occlusal events in some small herbivores. In P. M. Butler & K. A. Joysey, Eds. Development, Function and Evolution of Teeth, pp. 415–438. New York: Academic.

Robinson, M., Perry, D. M., Peterson, R. W., 1991, Optical Interferometry of Surfaces, *Scientific American,* 265, 66-71.

Solounias, N. & Moelleken, S. M. C., 1992, Tooth microwear analysis of Eotragus sansaniensis (Mammalia Ruminantia): one of the oldest known bovids, Journal of Vertebrate Palaeontology, 12, 113–121.

Solounias, N., Teaford, M. F., Walker, A., 1988, Interpreting the diet of extinct ruminants: The case of a non-browsing giraffid, *Paleobiology,* 14, 287-300.

Teaford, M. F., 1985, Molar microwear and diet in the genus *Cebus, American Anthropologist,* 66, 363–370.

Teaford, M. F., 1986, Dental microwear and diet in two species of *Colobus*. In *Proc 10th Annual International Primatology Conference*. *Primate Ecology and Conservation* (Eds. Else J, Lee P). Cambridge. University Press, Cambridge 63–66.

Teaford, M. F., 1994, Dental Microwear and Dental Function, *Evolutionary Anthropology,* 3(1), 17-30.

Teaford, M. F. and Glander, K. E., 1991, Dental microwear in live, wild-trapped Alouatta palliata from Costa Rica, *American Journal of Physical Anthropology*, 85, 313–319.

Teaford, M. F., Walker, A.C., 1984, Quantitative differences in dental microwear between primate species with different diets and a comment on the presumed diet of Sivapithecus, *American Journal of Physical Anthropology*, 64, 191-200.

Ungar, P.S., 1995, A semi-automated image analysis procedure for the quantification of dental microwear II, *Scanning,* 17, 57–59.

Ungar, P.S., 1996, Dental microwear of European Miocene catarrhines: evidence for diets and tooth use. *Journal of Human Evolution,* 31, 335-366.

Ungar, P.S., Simon, J-C and Cooper, J. W., 1991, A semiautomated image analysis procedure for the quantification of dental microwear, *Scanning Microsc,* 13, 31-36.

Ungar, P. S., Teaford, M. F., Glander, K. E. and Pastor, R. F., 1995, Dust accumulation in the canopy: a potential cause of dental microwear in primates, *American Journal of Physical Anthropology*, 97, 93–99.

Ungar, P. S. and Williamson, M., 2000, Exploring the effects of tooth wear on functional morphology: a preliminary study using dental topographic analysis, *Palaeontol Electron,* 3, 18.

Ungar, P. S., Brown, C. A., Bergstrom, T. S., Walker, A., 2003, Quantification of Dental Microwear by Tandem Scanning Confocal Microscopy and Scale-Sensitive Fractal Analyses, *Scanning,* 25, 185–193.

Walker, A. C., 1981, Dietary hypotheses and human evolution, *Phil Trans R Soc Lond B,* 292, 57–64.

Walker, A., and Teaford, M., 1989, Inferences from quantitative analysis of dental microwear. *Folia Primatologica,* 53(1-4), 177-89.

Walker, A.C., Hoeck, H., and Perez, L., 1978, Microwear of mammalian teeth as indicator of diet, *Science,* 201, 908-910.

Walker, P.L., Bernstein, S.A. and Gordon, K.D., 1987, An image processing system for the quantitative analysis of dental microwear, *American Journal of Physical Anthropology*, 72, 267.

Walker, P. L. and Hagen, E. H.,1994, A topographical approach to dental microwear analysis. *American Journal of Physical Anthropology Suppl*, 18, 203.

Whitehouse, D. J., 1994, *Handbook of surface metrology*. Philadelphia: Institute of Physics Pub.