DENTAL MICROWEAR ANALYSIS: DEVELOPMENT OF METHOD, AND INTERPRETATION OF ITS SIGNIFICANCE IN DIETARY AND FUNCTIONAL INFERENCES WITH A NEOLITHIC HUMAN SAMPLE FROM TEPE GANJ DAREH

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MASTER OF SCIENCE

Dental Microwear Analysis: Development of Method, and Interpretation of its Significance in Dietary and Functional Inferences.
C.L. Wang. Faculty of Dentistry, University of Manitoba, Winnipeg, Canada.

The use of this method in determination of human dietary and biomechanical differences has been rare. The problems researchers are faced with in utilizing this method have been mainly due to the amount of time and effort required to obtain significant results, the difficulties in obtaining a large sample size, and the lack of standardized methods used for analysis.

The present study has undertaken methodological and technical steps to reduce the amount of time required for completing a microwear analysis, at the same time providing a comprehensive standardized microwear analysis method that will give accurate results for future comparisons between different studies. A semi-automated image analysis system has been developed. This system has the ability for highly accurate recording of microwear feature dimensions, from an enhanced digital SEM image. A new method of microwear analysis, involving three key parameters that will account for all the important characteristics of a microwear pattern, is introduced.

These improvements in microwear analyses have been applied to the Neolithic Ganj Dareh human samples in an effort to determine their diet. Their microwear patterns are also compared with that of seven species of primates and other Mesolithic, Neolithic, and modern human samples obtained in other studies. The microwear pattern of Ganj Dareh individuals most resemble that of mesolithic or modern individuals.

Qualitative microwear analysis of two living individuals have been conducted. The objective was to assess the potential of utilizing microwear analysis for dietary and functional discrimination in modern individuals. Successive sampling of these individuals over the period of eight days allowed for estimation of the rate of turnover in microwear features in these subjects. The microwear pattern indicates highly destructive wear with a very slow rate of turnover, superimposed on teeth that show low levels of gross wear. This ambiguous pattern of wear may indicate that the microwear pattern, in these individuals, was not the result of normal functional wear, but may indicate infrequent catastrophic events, at the microwear level, that are indicative of parafunctional activities.

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ABSTRACT

Dental microwear analysis has been performed on many different groups of animals including primates, for the purpose of biomechanical and dietary determination. The use of this method in determination of human dietary and biomechanical differences has been rare. The problems researchers are faced with in utilizing this method have been mainly due to the amount of time and effort required to obtain significant results, the difficulties in obtaining a large sample size, and the lack of standardized methods used for analysis.

The present study has undertaken methodological and technical steps to reduce the amount of time required for completing a microwear analysis, at the same time providing a comprehensive standardized microwear analysis method that will give accurate results for future comparisons between different studies. A semi-automated image analysis system has been developed. This system has the ability for highly accurate recording of microwear feature dimensions, from an enhanced digital SEM image. A new method of microwear analysis, involving three key parameters that will account for all the important characteristics of a microwear pattern, is introduced.

These improvements in microwear analyses have been applied to the Neolithic Ganj Dareh human samples in an effort to determine their diet. Their microwear patterns are also compared with that of seven species of primates and other Mesolithic, Neolithic, and modern human samples obtained in other studies. The studies used for

comparison utilized different methods of microwear analysis, and the inadequacies of these studies have prevented accurate comparisons of results; the problems encountered and solutions required are discussed.

Qualitative microwear analysis of two living individuals have been conducted. The objective was to assess the potential of utilizing microwear analysis for dietary and functional discrimination in modern individuals. Successive sampling of these individuals over the period of eight days allowed for estimation of the rate of turnover in microwear features in these subjects. The microwear pattern indicates highly destructive wear with a very slow rate of turnover, superimposed on teeth that show low levels of gross wear. This ambiguous pattern of wear may indicate that the microwear pattern, in these individuals, was not the result of normal functional wear, but may indicate infrequent catastrophic events, at the microwear level, that are indicative of parafunctional activities.

1.0 INTRODUCTION

Dental microwear analysis has the ability to relate patterns of microwear features on the functional surface of teeth to dietary constituents and functional biomechanics of the masticatory apparatus. The major problems researchers face in conducting microwear analyses have been due to the tremendous amount of time and effort required to produce significant results, and the lack of standardized method used by researchers, causing an inability to compare results between different studies.

To address the first major problem, computer aided image analysis systems have been assessed in this study for their possible role in developing an automated microwear analysis system. Collecting and manipulating microwear data have traditionally been done by hand. Consequently this labour intensive process has occupied much of the researchers' valuable time. Any contribution in terms of automating part or all of the data collection and analysis of microwear features will mean an advancement in the efficiency of conducting microwear analyses.

To address the second problem, several new parameters for microwear analysis have been tested in an effort to produce a standardized microwear analysis method with the best power of discrimination possible. The basic rationale for choosing new parameters was that traditional methods of analysis have not considered all the possible significant characteristics of a microwear pattern. The development of such a method would allow for testing and comparison of results from different studies, without compromising the quality of these results.

Both traditional method and the newly developed approach have been used to analyze microwear data obtained from Tepe Ganj Dareh, a Neolithic site found in present day Iran. The results of the Ganj Dareh samples have been compared with results of other studies that included seven primate species, and human groups from prehistoric and modern samples. The significance of these findings have assisted determination of the mode of subsistence of the Ganj Dareh population. Since Tepe Ganj Dareh has been assumed to represent one of the earliest sites of sedentary food producers, the change from hunter-gatherer to agricultural subsistence may be accompanied by a significant change in dietary pattern. Therefore, the evidence from a dental microwear study can assist in answering questions of anthropological significance.

Qualitative microwear analysis have been conducted on two living adult humans from a fully industrialized population. The microwear analysis provided suggestion as to its potential in inferring modern diets, as well as functional, and parafunctional use of the masticatory apparatus. Successive sampling of the same individual allowed for an assessment of the rate of turnover in microwear features. Determination of the rate of turnover in microwear features provided an opportunity to estimate rate of tooth wear in a short period of time. The implications for such use in dental research have also been examined.

The goals of the present study were, 1) to develop an automated microwear analysis system, in order to improve the efficiency of these studies; 2) to develop a standardized method of microwear analysis that has the potential of having the best discriminative power between different microwear patterns; 3) to analyze the microwear pattern of the Ganj Dareh individuals, and infer their possible diet; 4) to examine the microwear patterns of living human subjects for potential uses of microwear analysis in dental research.

2.0 SAMPLE OF INTEREST: TEPE GANJ DAREH

2.1 Site description of Tepe Ganj Dareh

Tepe Ganj Dareh, or "Mound of the Treasure Valley", is a small mound site located in an upland valley of the Zagros mountains in Western Iran. The location of the site falls near the borders of the contemporary Kurdistan and Luristan, approximately 37 kilometres east of the city of Kermanshah and 10 kilometres west of the town of Hasin (Fig. 1a, 1b). At an altitude of approximately 1,400 metres, the valley is surrounded by mountains rising to over 2,000 metres (Agelarakis 1989; Meiklejohn 1980; Smith 1975, 1978; Smith and Mortensen 1980; Waddell 1994)[1].

The present mound is approximately 40 metres in diameter and covers roughly 1400 square metres. It rises to a height of about 6 metres above the surrounding valley floor, with an additional 2 metres of deposits below the surface (Smith 1978). A sample of roughly 20% of the mound was excavated by P. E. L. Smith over four field seasons; 1967, 1968, 1971, and 1974 (Fig. 2).

The site is divided into five distinct levels of occupation, designated A to E from the surface (Smith 1975). Radiocarbon dating findings suggest that Level E was occupied around 8,450 B.C., with the subsequent levels occupied most likely in the 8th millennium B.C. (Smith 1978).

2.2 Archaeological significance of Ganj Dareh

The site is of archaeological significance for a number of reasons. Its occupants showed significant architectural advancements; from open air fire pits with no buildings

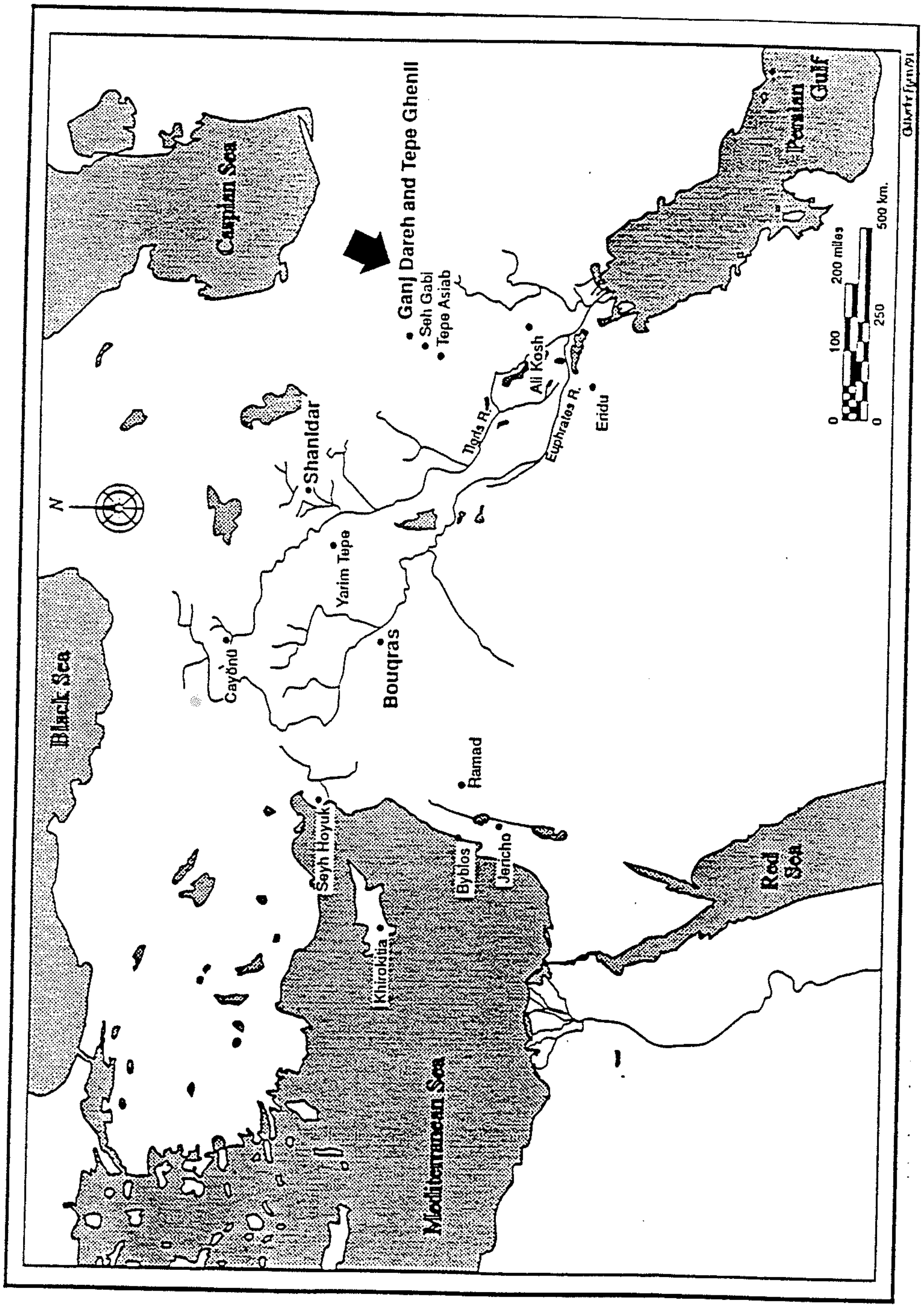
[1] All subsequent references to Smith will refer PEL Smith unless otherwise specified.

Figure 1. Maps locating Tepe Ganj Dareh and surrounding area.

Figure 1a. General location of Tepe Ganj Dareh in the Near East.

Figure 1b. Location of Tepe Ganj Dareh near the town of Kermanshah in the Zagros Mountains of Western Iran.

Figure 1a. Map locating Ganj Dareh in the Near East.



(Reprint from: Meiklejohn C, Agelarakis A, Akkermans PA, Smith PEL, and Solecki R. Possible origin of artificial cranial deformation in the Proto-Neolithic and Neolithic Near East: Evidence from four sites. Palēorient; 1992; 18(2):83-97.)

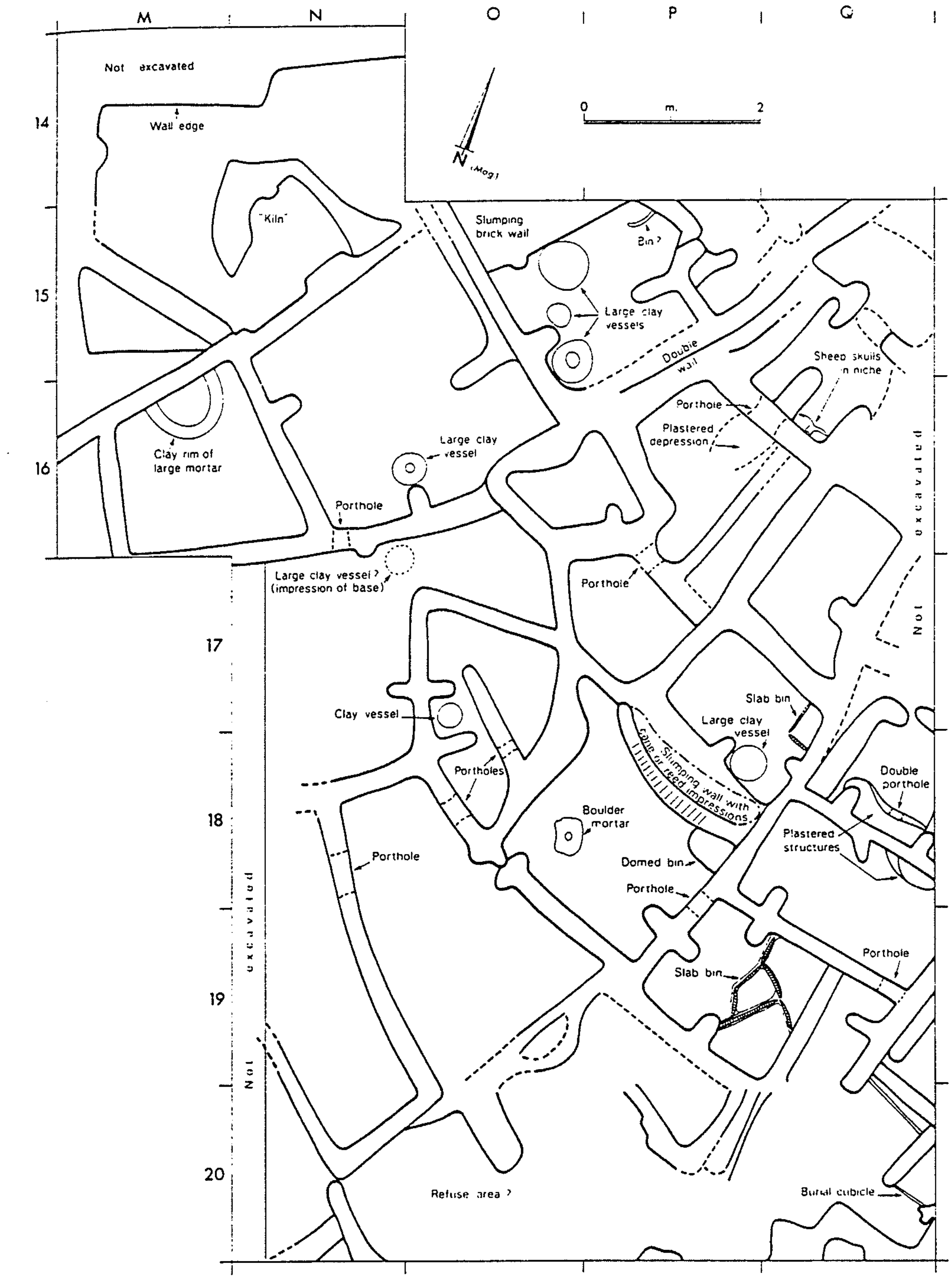
Figure 1b. Map locating Ganj Dareh in the Zagros Mountains of Western Iran.

(Reprint from: Smith PEL; Mortensen P. Three new "Early Neolithic" sites in Western Iran. Archives of Neolithic Near East; 1980; 21(4):511-512.)

Figure 2. Map of area excavated at Tepe Ganj Dareh.

Aerial map of the area excavated at Tepe Ganj Dareh. The Total area of the Ganj Dareh site was approximately 40 metres in diameter and covers roughly 1,400 square metres. A sample of roughly 20% of the mound was excavated, and is shown in the present map.

Figure 2. Map of area excavated at Tepe Ganj Dareh.



(Reprint from: Smith PEL. Architectural innovation and experimentation at Ganj Dareh, Iran. World Archaeology; 1990; 21(3):323-335.)

or shelters at Level E, to extensive use of clay in the form of brick and mud-walling, with increasing sophistication in the subsequent levels (Fig. 3). Some of the houses appear to have been two stories; many had small bin-like cubicles built in on the lower floors (Fig. 4). Large ceramic storage vessels were found in place inside some of the rooms as well.

A single pottery shard, and several clay "Venus" figurines, were discovered in Level E. These appear to be the earliest ceramic remains form the eastern arc of the fertile crescent. A number of kilns have also been identified in post Level D deposits, indicating prolific local manufacturing of ceramics (Smith 1978).

An elaborate necklace of stone beads and shells was found accompanying an adolescent burial in Level D (Fig. 5). Five of the shells were identified as Oliva, with its source being the Persian Gulf or the Indian Ocean. This finding represent the only evidence of distant contact at Ganj Dareh.

Faunal analysis revealed a wide variety of mammals, avifauna, reptiles, fish and invertebrates (Hesse 1978). Analysis of the remains suggest an overwhelming reliance on goat and sheep, in a controlled harvesting method; indicative of nomadic pastoralists. However, hunting was also important as shown by the presence of wild pigs, red and fallow deer, gazelles, ducks and pigeons, among others.

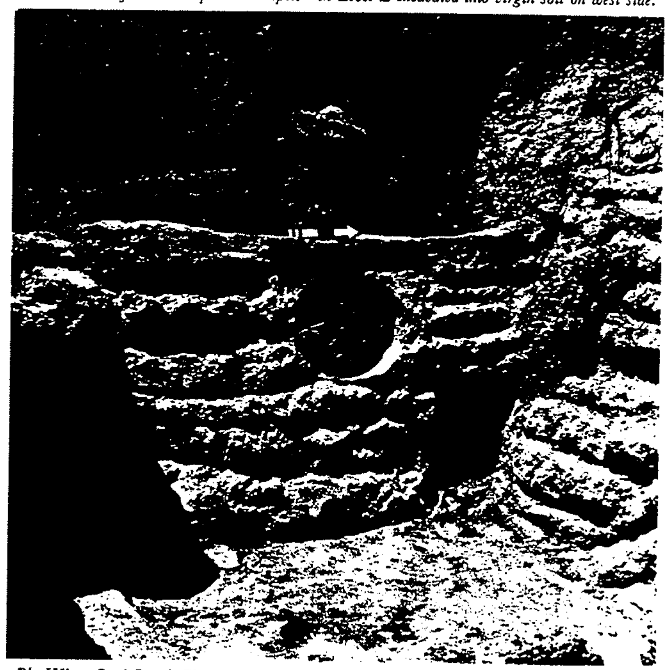
Floral analysis (Van Zeist, Smith, Palfenier-Vegter, Swijn, Casparie, 1986) indicates that both domesticated and non-domesticated barley were present in all levels of Ganj Dareh; although no other cereal was found. Wild lentils, almonds and a wide variety of seasonally available vegetable resources were also in evidence.

Figure 3. Technical innovations found at Tepe Ganj Dareh.

Technical innovations found at Ganj Dareh included the use of "firepits" as sites of cooking, and permanent structures constructed with the use of sun-dried mud bricks.

Figure 3. Technical innovations found at Tepe Ganj Dareh.

Pl. IIIa. Ganj Dareh Tepe. "Firepits" in Level E excavated into virgin soil on west side.



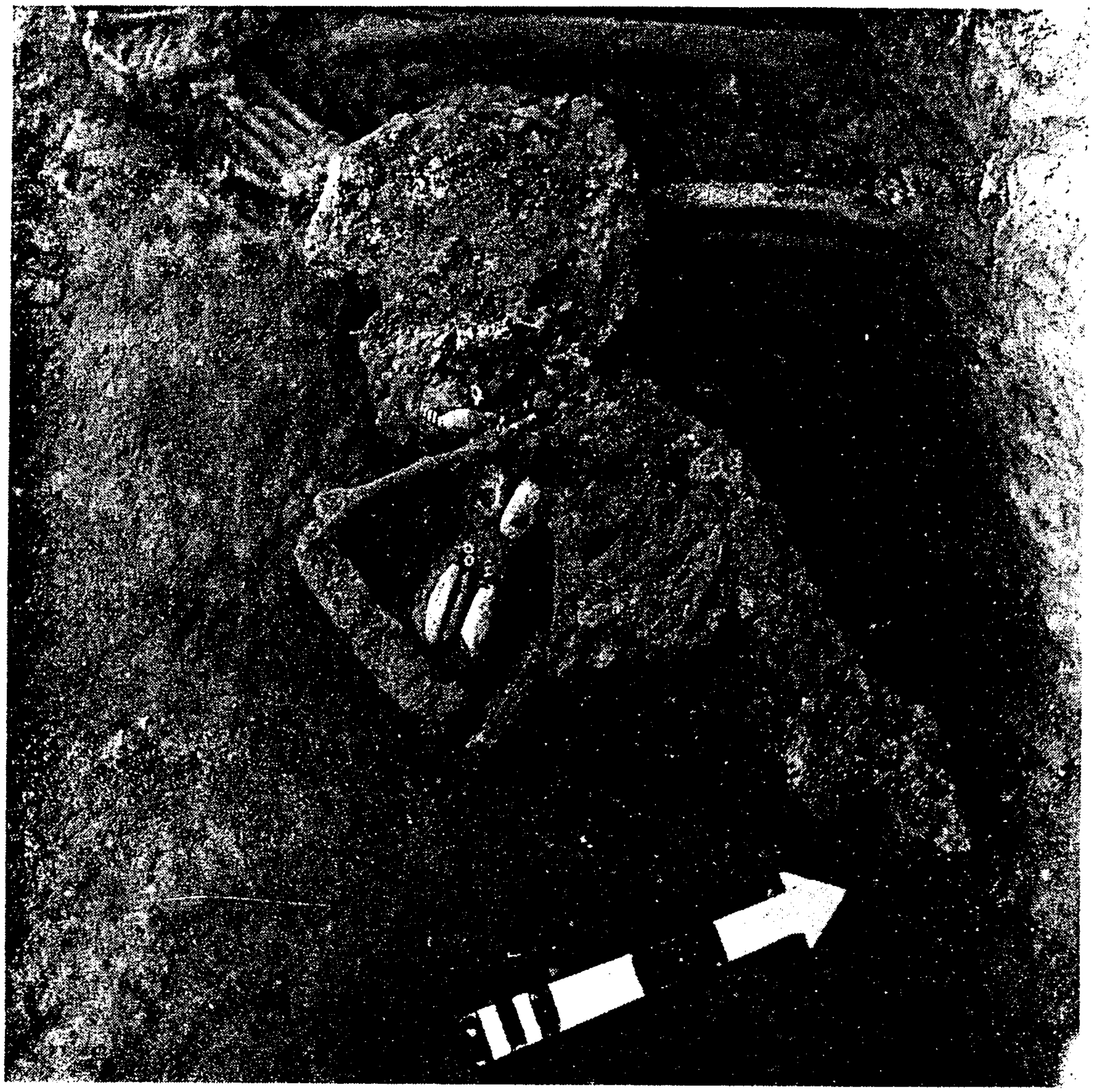
Pl. IIIb. Ganj Dareh Tepe. Brick wall of Level D structure, showing "porthole" aperture after unsealing.

(Reprint from: Smith PEL. Ganj Dareh Tepe. Journal of Persian Studies; 1975; 13:179-182.)

Figure 4. Multilevel construction used at Tepe Ganj Dareh

(Reprint from: Smith PEL. Architectural innovation and experimentation at Ganj Dareh, Iran. World Archaeology; 1990; 21(3):323-335.)

Figure 5. Child burial found at Tepe Ganj Dareh. A necklace made of sea shells was found to accompany this particular burial



(Reprint from: Smith PEL. Ganj Dareh Tepe. Journal of Persian Studies; 1975; 13:179-182.)

From the diachronic analysis of Ganj Dareh, it appeared to be a growing community at Ganj Dareh during the time of occupation, both in size and in sophistication. The site appeared to show a developing degree of control over its resources, with a move towards domestication of some plants and animals. Large storage containers and the presence of many mortars, pestles, and rubbing stones all suggest some form of food processing and storage. The finding of seasonal avifauna remains and use of building techniques, suggest the site is more than just a seasonal site of occupation. In Level E, where no solid architecture is found, winter occupations of the site is documented by the presence of seasonally migratory waterfowl as part of their dietary subsistence. During this time, the occupants may be in an early phase of pastoralism, where herders spent the winters here and moved up to higher pastures in the summers. Post-Level E deposits indicate at least part time summer occupation by the use of sun-dried bricks which can be made locally only between May and October. The abundance of bones of house mice from Level D onward may also reflect more sedentary conditions. Despite analysis of evidences from various sources mentioned above, there is still no clear agreement on the exact system of settlement and mode of subsistence at Ganj Dareh. Agelarakis (1989) contends the inhabitants were more or less sedentary. However, evidence forwarded by a number of authors (Hesse 1978; Smith 1975; 1984; Van Zeist *et al.* 1986) suggests the inhabitants of Tepe Ganj Dareh may have been semi nomadic pastoralists and that the site may not have been occupied on a year round basis.

2.3 The Ganj Dareh Skeletal Remains

Even with only one-fifth to one-quarter of the site having been excavated to this point, the remains of at least fifty-three individuals have been recovered. This represents the largest reported early Neolithic sample from this flank of the fertile crescent (Meiklejohn and Lambert, 1980). Burials of adult individuals were in both flexed and extended position, while young infants were commonly buried in plastered niches under the floors of living structures (Smith 1975). Grave goods were rare, and have primarily been found with young individuals. Three extended burials were recovered from a single covered mud-brick sarcophagus. A fire that swept through level D produced taphonomic conditions that significantly augmented the preservation of some individuals in this level. Preservation of the skeleton is better than expected due to this fire, but it also altered the morphological and structural components of the skeletal segments, which had undergone a calcination process. The high temperatures reached during the fire also caused significant alteration to the dental units. The enamel of some teeth was fractured off the underlying dentinal layer through thermal expansion of the dentin. The enamel that remains has a brittle, glassy appearance.

From the total sample, individuals have been aged from newborn to at least 50 years of age. Different methods produce contradictory results as to the maximum age of older individuals (Meiklejohn, personal communications). Both males and females are present, though no female or infant remains are yet identified from level E. Early results suggest an average age of death of less than 30 (Meiklejohn and Lambert 1980). Work at clarifying the demographic profile is currently under way (Meiklejohn, personal communications).

2.4 Pathology

Individuals at Ganj Dareh appear to have enjoyed relatively good health (Meiklejohn and Lambert 1980). In fact, general pathology appears to be relatively minor in comparison to contemporary sites in the region. However, the mortality profile of the skeletal remains seems to suggest a pattern of early age of death.

> "The general impression is, however, that the paradoxical pairings of low longevity with low pathology is encountered much more frequently in gathering and hunting populations than in later urbanized groups. We can therefore briefly conclude that the demographic and health aspects of this early Neolithic population still mirrored earlier patterning" (Meiklejohn and Lambert 1980).

Although only three of the adult individuals' dentitions were affected by carious lesions on the interproximal root surfaces, examination of the dental remains suggest that the frequency of other dental pathologies were relatively high (Agelarakis 1989). Enamel hypoplasia was found on individuals in all age groups except for the perineonates; suggesting growth disturbances, childhood diseases, trauma, or malnutrition. Local dental irritation in the form of mechanical and/or pathological conditions were represented by enamel projections into or toward the furcations, hypercementosis, as well as supracervical calculus accumulations. General periodontal disease was found in ten out of the fifteen adults, in the form of alveolar bone resorption and loss of interdental septae. Five out of the fifteen adults that were excavated with intact dentition had bony support that revealed dental abscesses, for the most part affecting the posterior teeth.

The wear on the available dentition appears to be moderate to severe. The majority of the masticating surfaces displayed uneven occlusal wear patterns, possibly as a result of functional modification during the stress of mastication. This was more evident in the older individuals, as was parafuntional use of the dentition as tools for non-masticatory activities. Seven out of fifteen adults evaluated had severe wear patterns, while nine showed concave occlusal surfaces with patches of secondary reparative dentin, and four revealed oblique occlusal planes (Agelarakis 1989).

3.0 LITERATURE REVIEW

3.1 Creation of microwear features

Functional wear on human dentition can be seen over time as tooth to tooth or tooth-food-tooth contacts slowly wear away surface enamel, forming a pattern of wear facets. At a gross level, the formation of these facets represents culmination of repeated abrasion or attrition events over a long span of time. At the microscopic level, individual features, such as a scratch or a gouge, can be seen on the facets of the enamel surface. Each of these features is the result of a single abrasive contact that has removed some of the surface enamel. New features overlap old ones as wear continues and the enamel surface is reworked and reduced.

The study of dental microwear is the method by which we look at these individual microscopic features, and try to interpret the events that have taken place to cause their formation. Wear on the enamel surface can be attributed to attrition, abrasion, and erosion. Attrition is indicative of tooth to tooth contact, forming polished surfaces with sub-parallel striations which are along the axis of the movement of the tooth. Abrasion wear occurs during tooth-food-tooth contact, where hard particles in the food can produce rough or pitted surface wherever opposing tooth surfaces transmit shear or compressive muscular forces. Erosion represents chemical dissolution of tooth material. Surfaces that are repeatedly exposed to such insult will show signs of erosion. Attrition occurs in the shearing and grinding phases of mastication, where the teeth in contact move laterally to one another, whereas abrasion occurs more often during crushing by the cusps, or cusp to fossa, with force vectors more perpendicular to the surface. By looking at these various functional surfaces on the dentition, one can infer the amount of crushing, grinding and/or shearing an individual is doing, as well as the direction of forces that produced these features.

3.2 Evolution of dental microwear studies

The formal study of dental microwear has only been around for the last couple of decades, but the precise origin of these analyses is difficult to trace. Many anthropologists that studied teeth have noted microscopic scratches or micro-features that may have the potential usefulness in interpreting jaw movement and tooth use (Butler 1952, 1972, 1973; Mills 1955, 1963, 1967). In 1962, Dahlberg and Kinzey (1962) published a paper describing the microwear features from a sample of prehistoric human teeth, seen by the light optical microscope. The results suggested that careful inspection of the variation in dental micro-features could further shed light on dietary differences within and between species. This appears to be the first definitive work that relates dental microwear to dietary inferences. No other dental microwear study was published over the next ten years. In the 70's, LeJeune and Baron (1973), and Wallace (1974), each published papers suggesting inferences can be made between the orientation of the striations on teeth and jaw movements or tooth use. Again, these studies were limited by the use of light microscope at low magnification. Walker's paper was particularly interesting since observations were made on differences in incisor microwear and feeding behaviour in some primates.

The development of high resolution casting techniques and the use of the scanning electron microscope in the 60's and 70's were of critical importance to the usefulness of dental microwear studies. Boyde (Boyde 1967, 1969, 1971, 1976, 1979, 1981, 1984) in the 60's and 70's demonstrated the benefit of using the scanning electron microscope for high magnification viewing of dental structures; and the immense potential of this technique began to gain recognition. In 1975, Shkurkin Almquist, Pfeihofer, and Stoddard published the first paper suggesting that the use of the scanning electron microscope for detailed examination of the enamel surface may be used for dietary reconstruction. The development of high-resolution casting techniques (Barnes 1978; Pameijer and Stallard 1972) in the 70's, allowed for microwear studies to be conducted on living specimens, and museum specimens that would otherwise be inaccessible to the rigors of laboratory studies. The combination of these two techniques, with further refinements over the following decade, allowed for detailed studies of a wide range of living and fragile specimens to be conducted; thus creating access to a new field of exploration.

3.3 Incisor Microwear

When considering function of teeth, one would naturally look to mastication as being its main purpose; but upon closer examination, most of the animals and early humans use their anterior teeth for many other parafunctional activities, and mastication generally only play a small part in dental wear (Brace 1962; Molnar 1971). Incisor microwear studies, therefore, have concentrated mainly on distinguishing the different types of parafunctional activities, and perhaps food preparation. Ryan (1981) was the first to look at incisal microwear; by comparing the orientation, density, and form of microwear features at low magnification. Three different species of primates were examined, and the interpretation concentrated on interspecific differences in either the amount of extraneous grit present in their food source, or their use of incisors for leaf-stripping behaviour. Ryan (1979a, 1979b) also looked at human samples such as Eskimos and Native American Indians in which incisors were known to be frequently used as a tool. Many characteristic microwear features were identified with their specific task.

Other researchers (Davies 1984; Teaford 1983) began looking at the incisor microwear features of primates for patterns of use, comparing those with positive overjet to those with an underbite. Significant differences were found indicating specific use of the underbite for food gathering and/or processing.

Kelly (1986) produced a doctoral thesis comparing different incisor microwear patterns of ten primate species. Interestingly, he found that methods of food procurement tasks and/or the physical properties and contaminants of dietary items have a major impact on incisor microwear formation. In addition, Teaford and Oyen (1986a, b) conducted an incisor microwear study on live primates with controlled diet. They found that the animals with soft diet often scraped the food off their fingers with their incisors, where as the hard diet animals rarely used their incisors for mastication. Those animals on a soft diet showed significantly more incisor microwear than the hard diet animals. These experiments showed great potential for functional and parafunctional differentiation when incisor microwear technique is used, but they also illustrate the difficulties and sometimes perplexing results one would have to sort through to produce meaningful results.

3.4 Molar microwear

In contrast to anterior teeth, mammalian molars are generally used for chewing rather than parafunctional activities such as grooming, etc. At first, molar microwear studies compared patterns of wear only in a qualitative manner. Two important papers in the 70's boosted interest in molar microwear studies. First, Walker, Hoeck, and Perez (1978) showed that seasonal changes in diet of one species of hyrax produced marked changes in molar microwear pattern. Analysis of the faecal matter of the hyrax revealed that opaline phytolith content increased dramatically during the part of the year when the animals switched to grazing from browsing. It was concluded that the opaline phytoliths were the most likely cause of the heavy scratch pattern found on the molars during that part of the year.

Rensberger (1978) documented different patterns of molar microwear patterns in six genera of modern rodents with known diets. The significance of this study is that he was able to match the differences in microwear pattern to specific diets in these animals; thus correlating specific patterns of wear with specific cause of wear. This effectively demonstrated the potential for detecting tooth-food-tooth interactions with microwear pattern studies.

A host of qualitative microwear studies followed (Walker 1981; Teaford and Walker 1983; Taylor and Hannam 1987), showing differences in molar microwear between browsing and grazing animals, open-country and forest herbivores, diets of vertebrates vs. invertebrates, all with encouraging results. The magnitude of differences in microwear pattern in these studies was sufficient for qualitative studies to be successful. However, these studies raised questions as to the discriminatory ability of microwear studies when the differences are not sufficient for qualitative inspection. For finer dietary distinctions, a method must be developed that would categorically define significant differences. The only way for this method to stand up to the rigors of scientific scrutiny was to develop a way to quantify these differences, and to test for statistical differences.

In order to quantify things, in this case it is the microwear features that are of interest, we must be able to categorize features into specific groups, giving objective description to the features that we wish to quantify. As the features are placed in categories, the number of features in each category will represent a pattern for that sample; that pattern can then be statistically tested against other patterns for statistical significance. As researchers have noted, in the qualitative experiments, the most obvious visual differences between different patterns are: 1) feature shapes, described as striations, scratches, gouges, and pits, all relating to their length to width ratio and their absolute size; 2) feature density, which is simply the number of features found in a given area; 3) feature direction, that is, whether the features follow a certain pattern of alignment. Walker's early works on hyraxes and browsers vs. grazers (Walker 1981; Walker, Hoeck, and Perez 1978) were the first to estimate the quantities of microwear features. Although no real definitions were given for his categories, other than in general terms such as long striations or short pits, he did make certain critical observations about the density, amount, and direction of the microwear features that were present.

Gordon (Gordon KD 1982)[2] was the first to attempt to categorize microwear features. Her intent was to provide baseline data characterizing intraspecific variability; a variety of chimpanzee molars were used. In this study, Gordon attempted to categorize features into pits, striations, or gouges through length to width ratios, but due to the low resolution of features on her photomicrographs (120 to 130X magnification), she was not able to obtain the exact width measurements. A series of intraspecific differences were found between facet types and molar positions. She was able to postulate that these differences found within an individual were caused by masticatory mechanical differences. Gordon's work pointed out the need to consider masticatory differences, molar position, and facet type when comparing results of interspecific and intraspecific quantitative microwear patterns.

Although Gordon revolutionized microwear analysis by introducing a quantitative method for analysis, the way that a feature is categorized as a pit or a scratch was still unclear; that is, although she selected the features for a particular category according to their length to width ratios, the ratios she had chosen to represent each category were not specified. She also conducted her analysis with low power magnification (125X), where feature width cannot be accurately measured. Different operators may have subjectively different category selection for a given feature.

Teaford and Walker (1984) conducted the first comparative quantitative microwear study two years after Gordon's work. In this study, they made the first attempt at objectively categorizing features, by defining a pit as any feature with length

[2] All subsequent references to Gordon refer to Gordon KD unless otherwise specified.

to width ratio of less than 10. By using this method, several significant observations were made. First, significant differences were found between species when using average feature length and width measurements on crushing facets, but not on shearing facets. Secondly, the frequency of features was not useful in distinguishing statistically between facets for species. Lastly, no significant difference was found between upper and lower teeth of an individual. In this study, Teaford and Walker were able to measure the length and width of each feature more accurately because they had chosen to analyze microwear features by using high magnification photomicrographs (500X magnification). They used the length and width measurement to categorize features into pits and scratches based on Gordon's (1982) comment that most features fell into these two categories. They also found that the feature length and width for some samples were not normally distributed. The result was that they believed microwear features were indeed made up of two separate entities. Therefore, they elected to use the chi-square test to analyze their data.

Many researchers since have chosen different length to width ratios to define pits and scratches. Grine and Kay (1988), in his research on hominid microwear patterns, suggested using the ratio of 4 to separate pit from scratch. This ratio was first discussed in his work on *Australopithecus* and *Paranthropus* (Grine 1986), where he arbitrarily chose features to be pits or scratches, and later measured them, and found that the pits he had chosen all had a ratio of 4:1 or less. Hayek, Bernor, Solounias, and Steigerwald (1991), using a mathematical model, found that a pit as a non-directional circular scar was best defined by a length to width ratio of 2:1. The ratio derived was the result of mathematical definition in which a circular conic's asymptotic upper limit is 2:1. As the ratio rises above 2:1, the eccentricity values decreases, until it becomes zero when the ratio reaches 10:1; the feature is then unquestionably linear. Teaford and Walker (Teaford 1985; Teaford and Walker 1984) both used the ratio of 10:1 as the cut off point for pit or scratch discrimination. However, in their later work, the researchers adopted the ratio of 4:1 as their cut off point for pit and scratch discrimination, for the reason that they found short scratches are often categorized as pits, thus skewing their results (Teaford 1988a; Teaford and Glander 1991; Teaford and Runstad 1992).

The use of pit to scratch ratio for quantitative analysis appears to be a natural progression from qualitative evaluation of microwear, where the differences between microwear patterns were evaluated on the consistency of the shapes of features. As quantitative analysis is becoming more sophisticated, various researchers have began to explore other feature parameters for their discriminatory value (Grine and Kay 1988; Molleson and Jones 1991; Molleson, Jones, and Jones 1993; Solounias and Hayak 1994 in press; Solounias and Teaford 1988). Solounias and Teaford (1994 in press) used pit diameter and estimated pit area as variables in their analysis of grazer and browser microwear patterns; they also tested different categories of feature classifications. The results indicated that the use of three categories, pits (pit/scratch ratio of <4) scratches (ratio of 4 to 100) and gouges (ratio of >100), gave the best diet separation. The use of diameter of features as a variable, together with pits, scratches, and gouges, provided the greatest separation of grazers, browsers, and intermediate feeders. Molleson and Jones (1991) and Molleson et al. (1993) also used pit diameter and pit area as different parameters in their multivariate analysis. They found that all the parameters tested were useful; however, the inclusion of pit area considered together with pit frequency gave a clearer picture of the function of the hardness of food, since larger pit sizes result in less number of pits in a given area. The reduced number of pits in such a case may mistakenly be interpreted as resulting from softer diet; the pit area gave a truer representation of the effects of a hard diet.

As many researchers have indicated, dental microwear studies are extremely time consuming, requiring an enormous amount of input to produce a minuscule amount of output (Gordon 1988; Kay 1987; Unger 1990; Walker and Berstein 1987). The present study, for instance, required the collection of data from over 11,000 microwear features, each having three measured variables. Such a large amount of measurements represent a sample of only 23 teeth, from 9 different individuals. There is a critical need for automating the data collection process if larger sample sizes are expected. Some researchers (Teaford and Walker 1984; Unger 1990; Walker and Bernstein 1987) have attempted computerized image analysis, where features are identified and measured by the computer; they have only succeeded in developing a semi-automated form of image processing, where features still needed to be individually identified by the operator, and the measurements were calculated by the computer. Others, such as Grine and Kay (1988) and Kay (1987), have tried using the analysis of power spectra obtained from numerical Fourier transformation of digitized micrograph images. Significant results have been shown with this method; however, they were not readily comparable to the results derived from manual quantification procedures, and do not give direct information concerning the microwear features themselves. In some cases, it has been suggested that this method may not separate taxa as well as manual procedures (Grine and Kay 1988).

One of the high tech methods for imaging surface topography that shows great promise, and is yet unexplored by microwear researchers, are the laser-based scanning systems. Sadler (1993) describes this system as being able to collect surface data as x-y-z coordinates at the same time, providing stricter geometric integrity than computer-aided tomography (CT) systems. Computer software are commercially available from computer-aided design and manufacturing, that are able to process the x-y-z coordinate data of laser scans. This system has not been applied to microwear studies, but appears to be very promising for achieving the high-resolution digital data analysis that microwear studies require, with readily available software for image analysis and data manipulation.

3.5 Microwear in dietary reconstruction

A benefit of microwear study for dietary reconstruction is that, unlike other functional indicators, it is minimally affected by an animal's phylogenetic history. Microwear features have the advantage of being formed directly as a result of functional activity during the animal's life time, unlike other morphological traits such as tooth size or occlusal morphology, which are variables that are genetically predetermined.

Some early natural and laboratory experiments provided interest to the field but often produced premature conclusions regarding dietary habits; some were due to the limitations of their technique and lack of comparative data (Covert and Kay 1981; Peters 1982; Puech and Prone 1979; Ryan 1979a, 1979b). As previously mentioned (p.14), Walker (*et al.* 1979) produced a significant early piece of work in which two sympatric species of hyrax were examined for dietary differences. One species was known to switch from grazing to browsing during the dry season. Significant differences in microwear patterns were found between grazing and browsing activities, mainly due to the presence of large amounts of abrasive silica in the form of grass phytoliths in the diet of the grazers. The discovery of the role of phytoliths was the good fortune of these early researchers, for the phytoliths provided a drastically different pattern of microwear in otherwise very similar species of hyrax. This work provided several important clues to further microwear research for dietary reconstruction. They include: 1) microwear patterns can be correlated to a difference in diet of the similar species; 2) microwear patterns have a rapid turn over rate such that seasonal changes in diet can produce significant change in the microwear pattern; 3) the cause of such a change in the pattern, in some cases, may correlate to, and be deduced from, specific agents in the diet.

Later works verified the distinct browsing-grazing continuum found with the two species of hyrax. Larger grazers and browsers such as antelopes, rhinoceroses and giraffes (Fortelius 1985; Solounias and Teaford 1988; Walker 1981) also displayed similar browsing-grazing microwear pattern differentiation. This pattern of differentiation will probably hold true for all browsers and grazers, since phytoliths found in grass are harder than enamel of all the different species. The scratching effect of the phytoliths and polishing effect of leaves will hold true for different taxa.

The first quantitative analysis of dental microwear was done by Gordon (1982, 1984b). In this early quantitative work, Gordon was able to show differences in microwear pattern in different tooth facets and position of tooth within the same species or individual. The intraspecific study illustrated that individuals with the same diet may have different microwear patterns, depending on where in the mouth the sample is taken from. She also speculated that the difference in the microwear pattern is due to jaw mechanics during function; where different types of facets provide different functions in chewing, and different teeth are subjected to different force vectors according to their location in relation to muscles and joints. The need for standardized sampling, in particular, tooth position and facet type, within and between species is a very important factor in producing comparable work. It also indicate that the role of jaw mechanics may overshadow any dietary differences when comparing the microwear patterns in animals with very different jaw mechanics and/or tooth types (Gordon 1984a, 1984b, 1984c).

Teaford and Walker (1984) provided the first comparative quantitative study of microwear. The study was designed to provide a profile of microwear patterns in different species of primates with different hardness of diet. The researchers found that those species of primates that were primarily leaf eaters and those that were primarily hard fruit eaters had microwear patterns that were positioned at opposite ends of pit to scratch ratio distribution; the intermediate species (those that ate both leaves and hard fruit) were found in intermediate positions. With a profile of hard and soft object feeder continuum for primate microwear patterns, Teaford and Walker were at a position to interpolate the microwear patterns of other primates into this data set, in order to infer their dietary habits. An extinct hominoid from the Miocene period, *Sivapithecus*, was found to match the microwear pattern of the intermediate feeder, chimpanzee.

Sivapithecus was thus presumed to be a mixed leaf-fruit feeder. Teaford and Walker were also able to make observations on facet differences. They found that although the lengths and widths of features on phase II (crushing facets) could be distinguished statistically, the microwear on the phase I (shearing facets) cannot be distinguished between the two species. The frequency of features was not useful in distinguishing statistically between facets for either species. This observation correlated with Gordon's (1982) results, that microwear patterns from different types of facets are not directly comparable. They also concluded that phase II facets may be more discriminatory than phase I facets for primate studies.

Microwear analyses have been used to distinguish dietary differences between taxa as well (Taylor and Hannam 1987; Strait 1993). Strait (1993) compared faunivore, frugivore and folivore mammals, and found that the mean pit frequencies were significantly higher for hard-object feeders (both hard-object faunivores and frugivores), than for folivores. Hard-object faunivores consistently demonstrated higher pit frequencies than soft-object faunivores. Microwear feature density, one parameter that was found to be non-significant in a previous study (Teaford and Walker 1984) and found only to be significant with age in another study (Gordon 1984b), appears to be higher in faunivores, thus providing separation of faunivores and frugivores; but unfortunately the differences are not consistently distinct statistically (Teaford 1985, 1988a). One important aspect of this study is that when metric analysis of gross molar morphology cannot distinguish between faunivores and folivores, microwear analysis of faunivores showed a significantly higher mean pit frequency than folivorous species. This study also illustrated some of the deficiencies of microwear analysis. In hard object feeders, despite differences in substance consumed, ie. hard insects versus hard bone, the mean pit frequencies are comparable, due to physical similarities of chitin and bone. Thus the specific dietary items may not be directly inferred by this method, only that the content is hard or soft; other sources of dietary information may be needed to construct the whole dietary picture.

Much of the works on primates that followed concentrated on problems such as; interspecific microwear differences in live animals where diets can be determined (Teaford and Glander 1991; Teaford and Runestad 1992); on closely related species with known diets (Teaford 1985, Teaford 1986); and on live primates of the same species with known seasonal or ecological differences in diet (Teaford and Robinson 1989). Each succession of studies enriched our knowledge of microwear pattern in primates and produced encouraging results that indicated microwear studies could be used to distinguish interspecific differences, intraspecific differences, and even seasonal or ecological differences. It also indicated that the microwear patterns differed more significantly as we move from intraspecific comparisons to interspecific comparisons when similar species are considered.

Microwear studies also continued on non-primate mammals. Molar and incisor microwear patterns have also been examined for large carnivores (Van Valkenburgh and Teaford, 1990; Robson and Young 1990), and ruminant species (Fortelius 1985; Solounias and Teaford 1988; Walker 1981). Researchers were able to find significant microwear differences between species within these broad categories and in each case extant species with known diets were used as controls to infer similar extinct species. Due to the large amount of work required to process a small amount of data, much of these data still lacked the volume that is required for extensive comparative purposes. It also appears that animals from one category cannot be directly compared to another, due to differences in tooth morphology and jaw mechanics. Base-line data are required for each category before significant comparisons can be made.

Microwear analysis on hominids have been very few, and those that were done have been evaluated on a qualitative bases only (Bullington 1988; Grine and Kay 1988; Pastor 1992; Puech, Albertini, and Serratice 1983; Ryan 1979a; Teaford 1989; Walker 1981). Researchers working with hominid material have only recently discovered the usefulness of this technique, and most of the initial problems have been due to the small sample sizes the researchers were faced with, since all were dealing with fossilized human teeth, where the samples are of great archaeological significance, but sample numbers are very limited.

The first comparative quantitative microwear study on hominids was done by Grine (1986), where he compared both phase I and phase II facets of *Australopithecus* and *Paranthropus*. He used both phase I and phase II facets for comparison, and found that both types of facets produced statistically significant differences between the genera. He was then able to use the phase II facet data, and placed them within the hard-object soft-object feeder continuum of Teaford and Walker (1984); both of these hominids were shown to be intermediate feeders, positioned on either sides of chimpanzee, with *Paranthropus* being closer to the hard-object feeder on the continuum. Due to the lack of any hominid base-line data, Grine had to resort to the use of primate data for comparison and diet interpretation. This piece of work still illustrated the potential use of microwear analysis in diet interpretation for hominids, and the need for future work on producing base-line data for comparison.

Molleson *et al.* (1991), and Molleson and Jones (1993) produced the first comparative quantitative microwear analysis on Neolithic human specimens. Her samples came from mesolithic, early neolithic, and modern sites found in the Middle East, and the 18th century Spitalfields collection. Comparisons were made between individuals who were known agriculturalists, subdivided into those that prepared their food extensively, ie. ground and cooked cereal, those that had not prepared the cereal, and those that were suspected to be hunter-gatherers. Although, her sample size was very small (2 to 5 teeth from each of 6 categories), she was able to find significant differences between hunter gatherers, primitive agriculturalists, and advanced agriculturalists, by using a multivariate analysis of variance. The parameters she used in her microwear analysis were: 1) total number of features; 2) pit density; 3) mean pit diameter and; 4) area in the field devoted to pits. Pit density and total number of features were parameters traditionally used for quantitative dental microwear comparisons. She cites previous works to explain the significance of pit density and pit size. Teaford and Walker (1984) found that increase in pit density reflects increased hardness of food. Ryan (Ryan 1979a, b) suggested that increase in pit size reflects an increase in the amount of crushing needed to comminute the food. Molleson introduced the use of area of the field devoted to pits as a parameter, because with its use, the relation between high pit density or large pit diameter and small number of observable features will not complicate the result. The parameter of total pit area will show the hardness, or amount of destruction, a diet will provide in the enamel. When total pit area and total number of features are considered together, a clearer picture of the nature of the diet can be inferred. High total pit area with low feature density indicates a very abrasive or destructive diet. Low total pit area with high feature density indicates a soft diet with attrition occurring to form the microwear features. Low total pit area with low feature density is only found with the weanling child in Molleson's study, where this pattern may suggest a soft diet with little attrition occurring. In this study, the pit size was significant in differentiating samples with hard or uncooked foods, from those with soft or cooked foods. Total number of features was found to be less in the young individuals; pit size and pit density was comparable to adults from the same group. The inference made is that pit size and pit diameter reflect the hardness of the food rather than the masticatory force generated in chewing. Overall similarity of the pit characteristics between groups consuming cooked foods suggest that meat eaters and vegetarians eating cooked food are not easily differentiated by their microwear feature parameters used in her study. With the help of faunal and plant remains, and available documented records in the case of the modern samples, the types of food ingested can be reconstructed. Therefore, this study concentrated on answering the question of what was done to the food before ingestion, and thus the extent of technological advances these populations possessed in the area of food preparation.

3.6 Complication with use of dental microwear for dietary interpretation

Artifacts on the surface of dentition can obscure or mimic "real" microwear features, ie. microwear features formed during the normal function of the dentition (Teaford 1988). In the case of fossilized teeth, these artifacts can occur both before and after the death of the animal.

In the living animal, there are three areas that should be examined to ensure that the microwear seen is the functional microwear of interest. First, functional microwear can be found on specific facets on the surface of the teeth. The formation of these facets are dictated by the way teeth come together and make contact, thus by the restraints of jaw mechanics. By examining the transition from a functional facet or surface to a non-functional surface, one should observe a drastic reduction in microwear features. Second, teeth will only begin to show meaningful microwear features after they have erupted into occlusion. Those teeth that have not erupted into occlusion, or do not have opposing teeth, can not be considered in microwear studies. Third, the microwear pattern on the chewing surface of teeth should occur in a somewhat regular pattern, since the formation of these features is ultimately dictated by the way teeth come together, thus by the jaw mechanics. Features that run in drastically different directions and those that change directions should be viewed with suspicion. The features present near the central groove of the occlusal surface will have greater degrees of freedom in direction, since they are formed by puncture-crushing at the end of phase I and beginning of phase II of the power stroke, where a greater change in jaw movements can occur. The most significant form of artifact in living animals is caused by tooth cleaning. The presence of the pellicle layer or saliva can obscure any microwear features if proper cleaning and drying procedures are not carried out.

With fossilized teeth, the possibility of post-mortem manipulation or damage can increase the presence of artifacts. The microwear features on these teeth should still follow the same pattern of normal functional microwear pattern unless the teeth have been damaged. The teeth should be examined for microwear-like features on non-functional surfaces. If the interproximal surface is available for examination, it will give the best indication of post-mortem wear. There may be an interproximal facet present, formed by tooth-to-tooth contact between neighbouring teeth. These facets show a very polished surface with very few microwear features, since they are not formed through function with abrasive food material, but only through the polishing effects of tooth-to-tooth contact. If any significant microwear features are found on these surfaces, it should serve as a good indication of post-mortem damage.

Post-mortem damage comes mainly from two sources: 1) those that occur after death of the animal but before fossilization; 2) those that occur during excavation and preparation of the fossilized material. The first type of damage would, at first glance, appear to be the most insidious. upon further consideration, one would discover that any effect such as chemical erosion or mechanical abrasion would occur over the whole tooth and the skeleton, not selectively on the functional surface of the tooth. Careful inspection of the condition of fossilization through the examination of all the surfaces of teeth and bones for indication of such destructive forces would reveal this type of artifacts. Even if it does occur on the occlusal surface of the tooth alone, it would probably consist of unusual sizes, shapes, and orientation (Gordon 1984b; Puech *et al.* 1983). The second type of damage, that occur during collection of samples, may have more potential to confuse the investigators. These types of damage can occur on selected surfaces, since it can be restricted to areas where researchers feel are important. Since tooth crown morphology is of critical importance to many areas of interest, they may receive more than their fair share of preparation. Two of the most common forms of damage comes from cleaning, and preservatives. The use of cleaning instruments, such as dental explorers, can cause unusually large gouges. Cleaning solutions such as acids can remove much of the surface details and expose enamel rod openings. A perfectly smooth surface with no enamel prism relief usually indicates the presence of preservative varnish on the surface. The presence of brush marks, or microwear features abruptly disappearing under a smooth surface, are additional tell-tale signs of application of a surface varnish. To date, there are no post-mortem factors that have been shown to mimic precisely normal microwear features of functional surfaces; with a sharp eye and some experience, one can detect signs of unwanted damage to the sample teeth.

Microwear patterns found on different molars of the same individual may be different, and can complicate microwear analysis. Gordon (1982) investigated the effect of molar position, facet type, sex and age on microwear pattern. She used the molar teeth from nine chimpanzees, all part of a museum collection. She found that although microwear pattern was not statistically different between first and second molars, it was different between first and third molar, as well as second and third molar. Moving from first molar to third molar, there appears to be a significant decrease in pit diameter but an increase in frequency, a decrease in striation length and frequency, and an increase in overall feature density. Gordon contributed these changes in microwear pattern with respect to tooth position as the result of the functional anatomy of the jaw itself. The relationship of molar position to the condyle and muscles of mastication dictates that molars that are further back in the tooth row will produce less shearing forces and greater crushing-grinding forces. The higher ratio of pits to scratches, decrease in striation length, and increased feature density found on phase II facets compared to phase I facets, are also due to the increase in vertical crushing-grinding forces that these facets are subjected to. Although there was not a significant difference in feature density between males and females, striations were found to be significantly shorter in females. There was also a significant decrease in feature density with increasing age, possibly due to reworking of features, causing less distinct delineation between features. The results of this study demonstrate some of the possible complications researchers are faced with if the sampling is not strictly controlled; at the same time further understanding of these complications will only assist us in designing better studies and aid in deciphering microwear results.

Doubts regarding dietary interpretation from microwear patterns have been raised due to lack of our present understanding of exactly how the microwear features are formed. There are two areas of research that should help us understand microwear feature formation a little better. The first area of interest is the effect of wear on different types and arrangements of enamel rods. Different types of enamel (prismatic vs non-prismatic), arrangement of enamel rods, and different sizes of enamel, all contribute to differences in resistance to wear (Covert and Kay 1981; Maas 1988, 1993; Strait 1993). The second area of interest is in the abrasive content of different foods. Rabinowicz (1965), a materials scientist, has noted that abrasive wear in pure metals is proportional to the hardness of the surfaces as long as the abrasive is harder than the abraded surface. However, Lipson (1967) showed that two minerals, chert and quartz, with identical hardness values, 7 on Moh's geological hardness scale, imposed differential degrees of abrasive wear when contacting steel. Chert, which is a tough mineral wore steel more than twice as fast as the much more brittle quartz. Ratner, Farberova, and Radyukevich (1967) reported that the relationship of relative breaking strength and strain to breakage is highly correlated to rates of abrasive wear in engineering polymers which are relatively homogeneous in hardness. Comparative studies of microwear (Covert and Kay 1981; Maas 1988, 1991; Peters 1982) suggest that a purely material property approach is over simplistic. Other factors such as exogenous grit, dental morphology and microstructure, and direction and intensity of chewing must also be considered. The research in this area is just beginning. Steps have been taken in specific areas of enamel morphology and materials properties, but the overall picture of the complex interaction of abrasives and enamel in living animals awaits future elucidation.

It is important to recognize that, although it may be a valuable and legitimate concern to study the specific causes of different microwear patterns, that information is not essential for dietary reconstruction, if different microwear states can be shown empirically to correspond to different dietary regimes. It is also important to understand that although dental microwear studies can reveal diet and jaw mechanic differences, one should use as many other sources of information as possible to reconstruct the whole picture (Smith HB 1984, Teaford 1988a).

3.7 Inferences about jaw mechanics from microwear studies

Microwear studies have also been used to furnish details about jaw movements and dental occlusion (Butler 1952, 1972, 1973; Gordon 1982, 1984a, 1984c; Mills 1955, 1963, 1967; Gingerich 1972, 1973, 1974; Every 1960, 1974; Teaford 1983; Teaford and Byrd 1989). Butler (1952, 1972) was able to show that the orientation of the wear facets and scratches revealed the type and degree of movement of the mandible. Mills (1955, 1963, 1967) found two distinct sets of scratches on wear facets, which he subdivided into two phases of jaw movements on the working-side. He further postulated that species differences in translation and rotation of the mandibular condyle can be separated by the differences in the relative lengths of the scratches formed by the different phases of jaw movement. These studies showed the orientation of jaw movements, but did not reveal the direction of movements. Gingerich (1972, 1973, 1974) reported evidence of an upward and backward mandibular movement during crushing-puncturing which he called orthal retraction; supported by the orientation of scratches on certain facets. Every (1960, 1974) found, in human dental patients, occurrences of jaw movements equal in orientation but opposite in direction to the movements of normal mastication. He postulated that the purpose of these movements are to resharpen the cusps and crests of teeth in response to blunting during normal use; a process he termed thegosis. He supported this theory with evidence of uniaxial appearance of scratches on thegotic surfaces, in contrast to multiaxial distribution on other wear surfaces.

Hiiemae and Kay (1973) and Kay and Hiiemae (1974) recorded jaw movement of several extant mammal species and did not observe either orthal retraction or thegosis. They did find an upward and anteromedial directed movement of the working-side mandible; which they renamed Phase I and Phase II. Since balancing-side contacts are occasionally found during jaw movements, they postulated that many if not all of the orthal retraction facets identified by Gingerich were actually caused by deviation of the jaw when these balancing-side contacts occurred. Rajaona, Woda, and LeJeune (1987) and Albuisson and Woda (1991) conducted microwear orientation studies on prehistoric and contemporary human samples, in which they found anterolateral movements of the jaw as well as anteromedial movements. These patterns of movements were explained by the variation in impact point when the teeth come into occlusion. To date, no behavioral study of living mammals has yet turned up firm evidence of either orthal retraction or thegosis.

Work on identifying the direction of jaw movement from microwear features has resulted in controversy (Ryan 1979b; Gordon 1984a, 1984c). Ryan (1979b) conducted an *in vitro* experiment on the shapes of artificially produced microwear features with predetermined magnitude and direction of force. Occasionally, these features showed asymmetry in shape which correlated with direction of movement producing them. Scratches had pitted or broadened ends at the point of initial contact, while tapered bodies narrowing away from the direction of movement were found. Gordon (1984a, 1984c) tested her sample of chimpanzee molars for indication of direction with this premise. Her results indicated a large variation in direction of jaw movement when evaluated by feature asymmetry. In fact, if feature asymmetry is a good indicator of direction, many cases showed that jaw movement occurs in both buccal and lingual directions in most of her samples. Gordon speculated that perhaps the *in vitro* conditions that Ryan used did not simulate *in vivo* activities, citing occurrences of microwear features that are asymmetric in shape, but are quite dissimilar to the shape that Ryan has described. Overlapping features also complicate identification of asymmetric features. Other researchers (Grine 1981; Teaford and Walker 1983; Walker 1981) criticized Ryan's model by stating that the model is an over-simplification of *in vivo* activities, and that scratch asymmetry is more reflective of force concentration than of jaw movement. Gordon further cited works from glacial geology, where wear features formed on rock surfaces which have been traversed by ice flows having asymmetric feature shape that was in reverse to Ryan's model. The broadened end of the feature is often found corresponding to the last point of contact. Flint (1967) also found that the features formed on upstream facing slopes have blunt ends in the downstream direction, whereas those on downstream facing slopes have blunt ends in the upstream direction. He concluded that asymmetric striations are not very reliable indicators of glacial flow direction. With all the evidence presented to date, scratch asymmetry itself cannot be considered a reliable source of directional information.

Gordon (Gordon KR 1984) created *in vitro* microfracture patterns of dental microwear scratches, and observed characteristic fracture patterns that indicate directionality. Fracture geometry of brittle materials have been correlated with the direction of abrasion (Bowden, Brookes, and Hanwell 1964). Gordon demonstrated that when abraded, like other brittle materials such as glass, partial or nearly complete Hertzian fracture cones can be observed on dentin and enamel. The bases of these Hertzian cones face in the direction of travel of the abrasive particle whenever a fracture pattern was observable. The sides of the three dimensional cones form a chevron-like pattern, indicating the direction of travel of the abrasive particle. The formation of this chevron-like pattern is not dependent upon the loading strength of the abrasive, unlike Ryan's model. Unfortunately, this fracture pattern can only be seen in 5% to 50% of the scratches that Gordon observed; factors such as optics of the SEM, preparation techniques, and subsequent abrasion or erosion of the tooth surface, can render the microfracture pattern unrecognizable. This method can give an indication of direction for some of the scratches on a facet independent of loading strengths; but cannot be considered a reliable indicator of all, or even the majority, of the direction of scratches on a given facet.

Teaford (1983) attempted to find other indications of feature direction with the use of the molar teeth of guinea pigs. Since guinea pigs have continuous erupting molars, the occlusal surface of these teeth show both enamel and exposed dentin. Since dentin is softer than enamel, the wear on dentin is more severe than enamel, when exposed to the same source of abrasion. Teaford was able to show that during normal mastication, leading enamel edges shelter the dentin surfaces adjacent to them in such a way that the transition from enamel to dentin is quite smooth. At the trailing edges, however, dentin precedes enamel. Thus the dentin surface was worn more deeply at the trailing edge, producing a pronounced step at the transition from dentin to enamel (Teaford 1983). He went on to show that guinea pigs, due to limitation in jaw movements during chewing, have a fairly predictable power stroke, producing fairly regular orientation in their microwear features. When one of their trigeminal nuclei was damaged artificially, the damaged side showed significantly different microwear features, both in orientation and in feature size and pattern. No specific patterns of microwear features were identified with specific change in jaw coordination, only that a change in motor function can have a dramatic effect on microwear pattern.

Teaford's method of detecting directionality requires both enamel and exposed dentinal surfaces to be involved in the microwear features in order to identify the direction of those features, therefore, not all features' direction can be determined by this method. To this day, no reliable method has been developed that can predictably indicate the direction of all, or even most, of the features' direction on a given wear facet. Although the determination of features' direction can benefit the study of functional jaw mechanics, the method for such prediction is still lacking, and awaits further development.

3.8 Inferences about tooth wear from microwear studies

The rate of enamel wear in animals has been shown to correlate with the hardness of diet and age-related changes (Barrett 1958, Molnar 1971, Teaford and Oyen 1989). Teaford and Oyen (1989) conducted a longitudinal study of dental wear detection by observing the pattern of microwear features. Fifteen vervet monkeys, divided into two groups, were raised on hard vs. soft diets. Cusp heights were recorded at the beginning of the experiment. Results of this experiment showed that there was significantly greater wear in animals with harder diet. The greater wear was accompanied by a microwear pattern that is characterised by large pits. For both the hard diet and soft diet groups, the rate of wear was much greater than any published data for Western industrialized humans; cusp height reduction of 71 to 286μm per year was found in these animals. In a separate experiment Teaford and Tylenda (1991) went on to examine the rate of turnover of microwear features. By examining nine adult humans, they found that the rate of microwear turnover, thus overall wear, is much slower in humans than in laboratory monkeys raised on either hard or soft diets. The rate of turnover can be estimated by observing the number of new features that are formed over a short period of time (in this case, three days between observations), and extrapolating the amount of time required for complete turn over of features; in this experiment, the subjects would have a complete turnover of microwear features in 60 days. Crushing-grinding (phase II) facets were found to have a higher turnover rate than shearing (phase I) facets. Interestingly, the only exception to this pattern is an individual that ate mainly salads and fresh vegetables. Teaford postulated that vegetables required greater amount of cutting and shearing during chewing than grinding, with a correspondingly greater rate of wear on shearing facets. The results of this study was significant in that microwear studies may be utilized for examining relative change in dental structures over a very short period of time, and different patterns of chewing may demonstrate different wear rates on specific areas of a tooth.

Teaford and Glander (1991) conducted further studies that investigated the rate of microwear feature turnover on monkeys with known diets. The monkeys were identified as to the amount of shearing vs. grinding that was required by their natural diet. First, they found that the rate of wear can be different between shearing and crushing facets. The difference in the rate of wear on different facets was confirmed by specific types of diet that required different amount shearing and crushing. They were then able to postulate that the difference in wear between shearing and crushing facets of the same individual can be used to infer the kind of diet the individual consumed. The use of this method can compliment the inferences made by the study of microwear feature patterns found on a single facet.

4.0 METHODS

The methods used in this project to obtain quantitative data of tooth surface microwear were a modified version of those found in the literature (Barnes 1979; Beynon 1987; Gordon 1988; Rose 1983; Teaford and Oyen 1989a). The object of interest in this project is the set of microwear features present on enamel surfaces of human teeth. In order to quantify these microwear features, the enamel surfaces must be examined with the aid of the scanning electron microscope. Microwear features can be found on occlusal wear facets on teeth. They are formed when the enamel comes into repeated contact with abrasive objects. When features were located on the facet of interest, a photomicrograph was taken, and the features were then measured and counted on the photomicrographs. Statistical analysis was then applied to the quantified microwear features to determine a pattern of microwear formation, which can be compared to other patterns of wear formation.

In this archaeological sample, there were ten individuals that had one or more of their permanent first molars recovered. From these ten individuals, 31 first molars were available, but only 23 of them, from nine individuals, had identifiable facets that were amenable to the study; the other 8 were either too worn, or were not worn at all. Detailed relevant data for each of the individuals are presented in Table 1. From these 23 teeth, two replicas were made of each tooth, and two photomicrographs were taken from phase II chewing facet 9 of each of these replicas. When facet 9 was not present, facet 10n or X were substituted, recognizing that although they were located in another area of the tooth, they were still phase II chewing facets. All phase II facets are

Table 1. Ganj Dareh Sample Profile

Twenty-three first molars from nine individuals were available from the Ganj Dareh samples for the purpose of microwear analysis. The available information regarding these individuals are presented in this table.

The teeth numbering system is defined as the following:
16 refers to upper right first molar
26 refers to upper left first molar
36 refers to lower left first molar
46 refers to lower right first molar

There were a total of 9 individuals with 23 teeth.
-2 individuals with 4 molars available
-3 individuals with 3 molars available
-2 individuals with 2 molars available
-2 individuals with 1 molar available

-a total of 6 tooth 16 were available
-a total of 6 tooth 26 were available
-a total of 5 tooth 36 were available
-a total of 6 tooth 46 were available

-there are 7 pairs of teeth from 6 individuals available for upper to lower teeth comparison
-there are 8 pairs of teeth from 6 individuals available for right to left teeth comparison

Table 1. Ganj Dareh Sample Profile

Individual Identification Number	Teeth Available		Number of Teeth	Age	Sex
10	16	26	2	8-12	?
13			1	50+	Male
		36			
13a			1	?	?
		36			
15		26	3	15-20	Female?
	46	36			
16	16		2	6-8	?
	46				
17	16	26	4	15-19	Male
	46	36			
20	16	26	3	30-40	Male
	46				
23	16	26	3	20-25	Female
	46				
40	16	26	4	20-25	Male
	46	36			

formed during the crushing/grinding phase of the chewing cycle, when the buccal facing inclines on the lingual cusps of upper molars occlude against the lingual facing inclines on the buccal cusp of the lower molars. Therefore, the microwear patterns found on all phase II facets on the same tooth should be the same. The locations of the various facets are illustrated in Figure 6.

In addition, two volunteers were recruited from the student pool at the Faculty of Dentistry, University of Manitoba. Impressions of the two first molars on their dominant side (which they had identified as the side they most often chew with) were taken on two separate occasions, eight days apart. Casts were made of these teeth, and photomicrographs of facets on the occlusal surfaces were taken with the aid of the SEM. Examination of their microwear features was done on a qualitative basis.

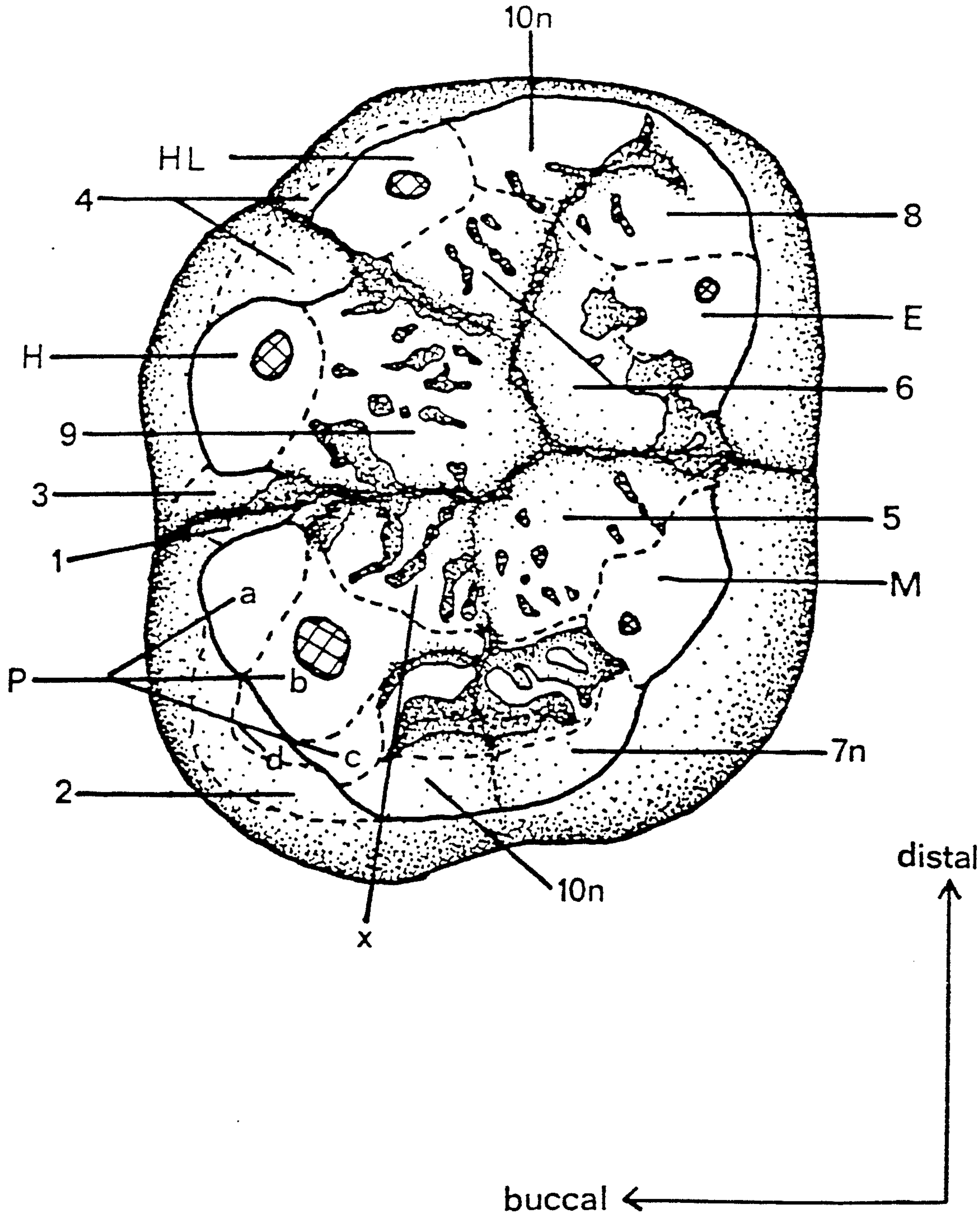
4.1 Specimen Replication

The specimens of interest was obtained from archaeological samples excavated from Ganj Dareh, presently held at the University of Winnipeg, as well as live human subjects recruited from the student pool at the Faculty of Dentistry, University of Manitoba. Microwear features of interest were found on the functional wear facets formed on the occlusal surface enamel of molar teeth. The enamel surface must be observed with the scanning electron microscope (SEM) to obtain photomicrographs. Human enamel can be successfully observed in the scanning electron microscope (Risnes and Stolen 1981), but the use of replicas, instead of the original specimens, has several advantages; 1) replicas can be easily transported and mounted for viewing without damage to the precious, and often delicate, original specimen; 2) the original specimens

Figure 6. Map of molar facet numbering system.

The molar facet numbering system used in this study was first proposed by Kay (1977) and later modified by Gordon (1982). Phase I shearing facets were found on the buccal facing inclines of the buccal and lingual cusps (1, 2, 3, 4, 5, 6, 7n, 8). Phase II crushing facets were found on the lingual facing inclines of the buccal cusps (9, 10n, X).

Figure 6. Map of molar facet numbering system.



(Reprint from: Gordon KD. A Study of Microwear on Chimpanzee Molars: Implication for Dental Microwear Analysis. American Journal of Physical Anthropology; 1982; 59: 195-215.)

are often too large to fit into the scanning electron microscope's chamber, especially if the teeth are still attached to the mandible or maxilla; 3) replicas can be made in the field or outside the laboratory setting; important when the specimen cannot be removed from its present location, such as museum pieces; 4) replicas can be made on living subjects; 5) replicas can be made on the same subject in succession for the purpose of longitudinal studies, and 6) replicas can be taken of specimens that cannot otherwise withstand the vacuum that is required for the operation of scanning electron microscopy.

4.2 Specimen cleaning procedures

Before the replication procedure can take place the specimens must be clean. With archaeological samples the specimen may be covered with a macroscopic and/or microscopic layer of dirt, organic debris, or preserving varnish. With living specimens, the teeth in the mouth may have food debris, or an organic pellicle layer present on the surface of the teeth. The specimen must be free of all debris before the impression is taken, or else the debris will also appear on the casts, obscuring valuable microwear features. A series of mechanical and chemical cleaning procedures were followed to ensure that both organic and inorganic contaminants were removed (Gordon 1988; Teaford 1989).

Cleaning of archaeological samples began by soaking the specimen in a 2.5% solution of sodium hypochlorite (bleach), to loosen any macroscopic dirt and grease. A clean water rinse, and if necessary, gentle brushing with a soft tooth brush will remove the surface dirt mechanically, and ensures that the bleach solution is rinsed away. Care must be taken when brushing the tooth surface, to ensure that any dirt being removed from the tooth surface does not scratch the enamel, causing feature artifacts. Whenever possible, tooth brushing was replaced with the use of ultrasonic cleaning. The tooth was emersed in a mild detergent specifically for ultrasonic cleaning, and was left in the ultrasonic cleaner for one to three minutes. When removed from the ultrasonic cleaner, there were no surface debris visible. The specimen was then rinsed once again to remove the detergent, followed by the application of acetone with a soft cotton gauze. The acetone should remove any organic film that was remaining on the tooth surface. The occlusal surface of the tooth was then wiped clean with alcohol, again applied with a soft clean cotton gauze. Finally, the entire specimen was air dried with compressed air from a commercial source, such as those used for removing dust from photographic equipment. Compressed air from mechanical pumps was not acceptable, mainly due to the oil contaminants that were often found in the conduits of these machines. The agents used for cleaning tooth specimens, including tooth brushes, were used in different combinations, by previous researchers, with no noticeable effect on the dental microwear patterns (Barnes 1978; Gordon 1982; Teaford and Oyen 1978; Rose 1983). Photomicrographs of original tooth surface taken before and after following the above cleaning procedures are illustrated in Figure 7.

To clean the tooth surfaces of live subjects, the subjects were asked to brush their teeth for two minutes with commercial tooth paste. The tooth of interest was then subjected to light cleaning with a slow speed rubber cup, while being rinsed with water. Since live subjects will not have any macroscopic debris on their teeth after the tooth brushing, cleaning with the rubber cup was only an attempt to remove as much of the

Figure 7. Comparison of tooth surface before and after cleaning.

The surface of archaeologically recovered dentitions were often covered with debris that may have obscured microwear features of importance. The source of surface debris may have been dirt from the original burial site, or from varnish placed onto the surface as a preservative by the curator of the material.

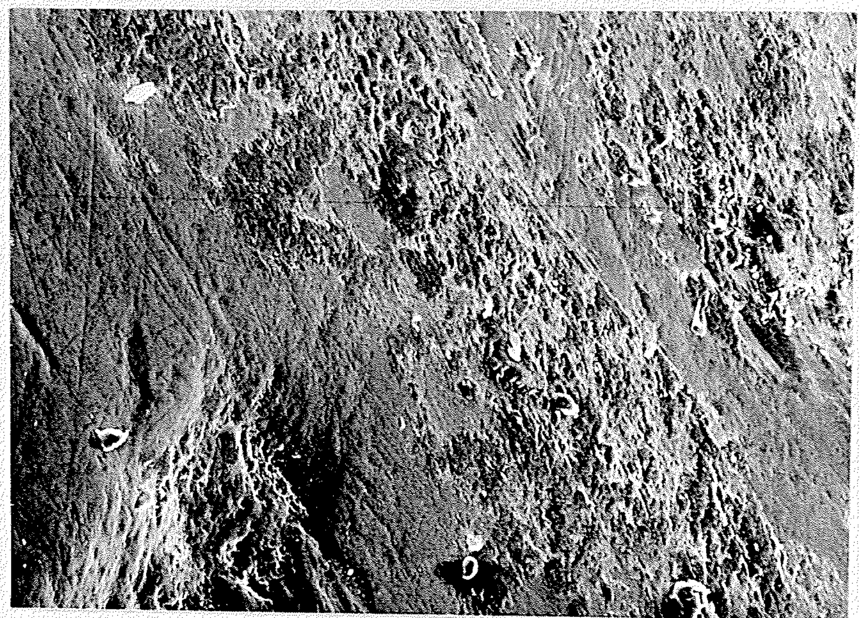
The tooth surface was cleaned before replication, to ensure proper duplication of all the microwear details. The teeth were first cleaned with a 2% bleach solution, then placed into an ultrasonic cleaner for 1 to 3 minutes, followed by an acetone and alcohol rinses. The teeth were then wiped and dried.

The cleaning procedure used for this study was suggested by Gordon (1988). The cleaning procedure had been designed to remove surface debris without causing damage to the tooth surface. Tooth 37 (lower left second molar) from individual 20 of the Ganj Dareh samples was used to test the cleaning procedure. SEM photomicrographs were taken of the tooth surface at 480X magnification.

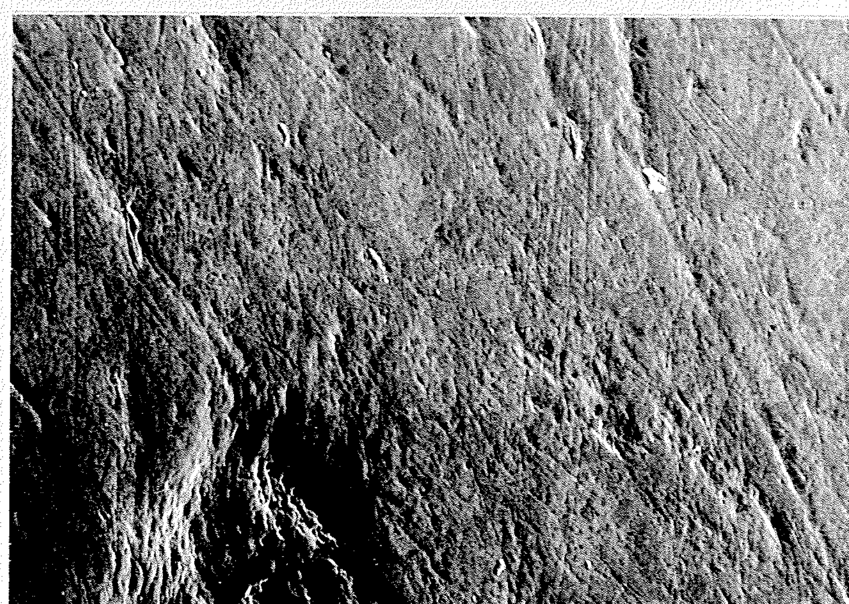
Figure 7a. Tooth surface before cleaning procedure. Notice the amount of surface debris present. In this case, the debris was most likely from the original archaeological site.

Figure 7b. Tooth surface after cleaning procedure. Microwear feature can now be easily identified with very little obstruction from debris.

Figure 7. Comparison of Tooth Surface Before and After Cleaning



a Before



b After

pellicle layer as possible without any chemical or abrasive agents that might alter the microwear pattern. The tooth was then dried with an air syringe, and kept dry by cotton rolls, in an attempt to isolate the tooth from saliva contamination.

4.3 Impression technique

A number of commercially available impression materials used for dental purposes have been used in the past for this purpose (Barnes 1979; Beynon 1987; Gordon 1988; Rose 1983; Teaford and Oyen 1989b). The requirements for making a good impression for microwear studies are; 1) ability to produce high resolution of details, at least better than 1 micron of differentiation; 2) compatibility with casting material, in this case, low viscosity epoxy such as those used as specimen embedding material for electron microscopy as used in this study; 3) dimensional stability, preferably for weeks or months, since the cast may not be poured for days if they are taken away from the laboratory. Of the many choices of commercially available dental impression materials, polyvinyl siloxane impression materials appear to fulfil these requirements the best. Polysulphide rubber base materials produce high resolution impressions, but are incompatible with epoxy casting materials. Polyether rubber does not produce as detailed impressions, and is also incompatible with some casting material. Hydrocolloid impressions are not dimensionally stable for more than a few minutes.

Of the polyvinyl siloxanes, the addition-reaction type have better long term stability than the condensation-reaction type. Condensation-reaction type of silicone eliminates ethanol as a byproduct of polymerization, and thus contributes to higher polymerization shrinkage. Addition-reaction polyvinyl siloxanes are found to give highly detailed impressions, and often capable of producing impressions of greater than 0.1 micron resolution (Barnes 1979). They are also compatible with epoxy casting materials, and are dimensionally stable up to several weeks or months when proper storage conditions are available. Many brands of commercially available polyvinyl siloxanes have been used successfully for this purpose; products such as Xantopren Blue, CutterSil Light, Express, Reprosil, President, among others have the resolution and stability capabilities required, and were reported in previous literature (Barnes 1979; Beynon 1987; Gordon 1988; Rose 1983). The major problem with choosing the best product is that the products are constantly being reformulated by the manufacturers, since the need for duplication of surface detail at the microwear level is not generally a priority, and is often compromised when ease of use and viscosity, among other practical considerations are being sought.

A couple of the recommended materials were tested to ensure the quality of the impressions was not compromised by manufacturer reformulation. The brands chosen, Reprosil (L.D. Caulk) and President Jet Light body (Coltene), were recommended by Dr. Mark Teaford (personal communications), one of the leading microwear analysis researchers. Dr. Teaford suggested that often the right impression material is found through trial and error, since there are no manufacturer's data on resolution of replication at this magnification. Twenty Epon epoxy replicas were made for Reprosil and President impressions, and were examined with the SEM. Photomicrographs of the replicas were compared to the photomicrographs of the original, uncoated, tooth (Figure 8).

Qualitative evaluation shows that the President/Epon combination appears to produce superior detail, when compared to the Reprosil/Epon combination. Reprosil, an impression material that was successfully used for previous studies, has recently changed their formulation, making it easier to mix, but obviously by diminishing its detail reproduction at higher magnification.

By examining the negative impressions themselves with the SEM, one can eliminate an additional step of making a positive cast; presumably eliminating the inaccuracies that might accompany such an additional step. A thorough investigation of the literature revealed that a few researchers have attempted such a technique (Galil and Gwennett 1975). In general, however, direct examination of the primary impression is problematical for two reasons. First, when gold is coated directly into the impressions, it may craze. It has been suggested that this may be due to the distortion of the silicone rubber, owing to its high coefficient of thermal expansion (Grundy 1971). As part of the preliminary testing of materials, ten polysulphide and silicone negative impressions were coated with gold/palladium, and viewed with the scanning electron microscope (Fig. 9). Craze lines of a consistent pattern can be seen in all of the samples, although it may not affect all areas of a given sample, obscuring the details of the impression (Fig. 10). Second, interpretation of the primary impression is difficult. It takes a great deal of effort to accustom oneself to reading of the surface details, making interpretation of features difficult. The negative impression also makes viewing difficult, since the impression is shaped like a cup, and the rim of the "cup" casts a shadow over the surface of interest, reducing the contrast of photomicrographs taken.

Figure 8. Comparison of Replicas to Original Tooth Surface.

Original tooth material can be placed in, and viewed directly with, the scanning electron microscope. Original tooth material may not always be available for such purposes. Gold-palladium coated negative impression can also be viewed with the SEM. Unfortunately, surface contrast was not sufficient to reveal details, and the gold-palladium coating often crazed when the impression material under went thermal contraction in the vacuum chamber of the SEM. Recent reformulation of Reprosil polyvinyl siloxane impression material produced poor resolution of details when casts were made from these impressions. President Jet polyvinyl siloxane impressions produced casts with adequate detail resolution for dental microwear analysis.

Tooth 47 (lower right second molar) from individual 497 of the Ganj Dareh samples was used to conduct comparison tests of tooth surface viewed directly with the SEM, to and surfaces of the impressions and casts made from it.

Figure 8a. SEM photomicrograph taken at 200X magnification of the original tooth surface.

Figure 8b. SEM photomicrograph taken at 200X magnification of the negative impressions made with Reprosil. The area included in this photomicrograph is similar to that which is shown in Fig. 8a. Notice the craze lines found uniformly on the surface of the impression.

Figure 8c. SEM photomicrograph taken at 200X magnification of the Epon epoxy replica made from President Jet impressions. The area included in this photomicrograph is similar to that which is shown in Fig. 8a. Notice the surface detail that has been accurately reproduced by this method when compared to the original.

Figure 8d. SEM photomicrograph taken at 480X magnification of the original tooth surface.

Figure 8e. SEM photomicrograph taken at 480X magnification of the Epon epoxy replica made from Reprosil impressions. The area included in this photomicrograph is similar to that which is shown in Fig. 8d. Notice the insufficient surface detail on this cast made by this method.

Figure 8f. SEM photomicrograph taken at 480X magnification of the Epon epoxy replicas made from President Jet impressions. The area included in this photomicrograph is similar to that which is shown in Fig. 8d. Notice the surface detail that has been accurately reproduced by this method when compared to the original.

Figure 8. Comparison of Replicas to Original Tooth Surface.

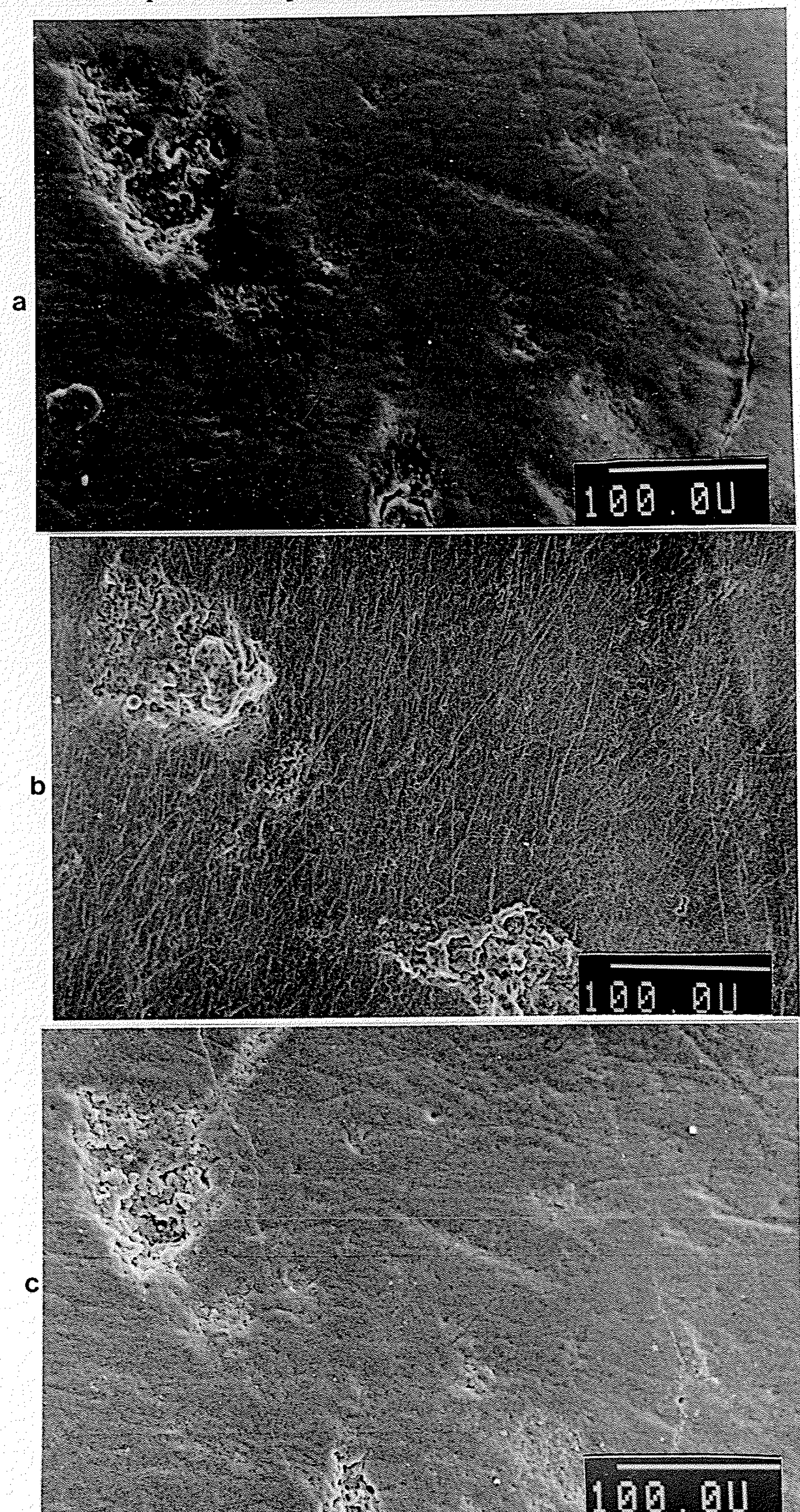
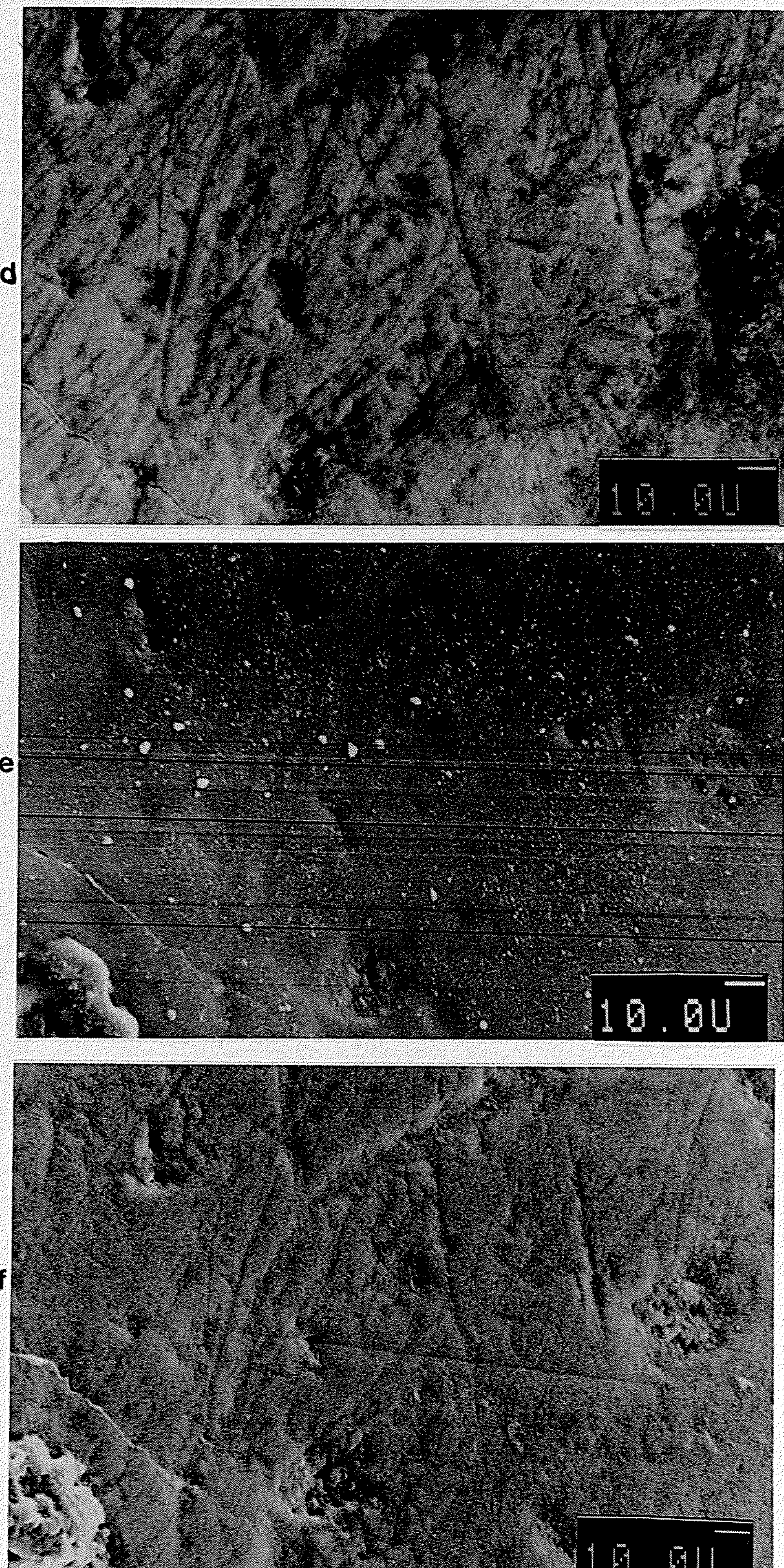


Figure 8. Comparison of Replicas to Original Tooth Surface.



d

e

f

Figure 9. Cast and Impression mounted and plated for SEM viewing.

Cast and impression from tooth 47 (lower right second molar) of Ganj Dareh individual 497 were mounted on SEM stubs and coated with gold-palladium for conductivity. The cast (a) and impression (b) were trimmed to fit flush and as close as possible to the SEM stub. The occlusal surface paralleled the stub table surface, to produce accurate orientation of the occlusal surface when viewed with the SEM.

Figure 9. Cast and Impression mounted and plated

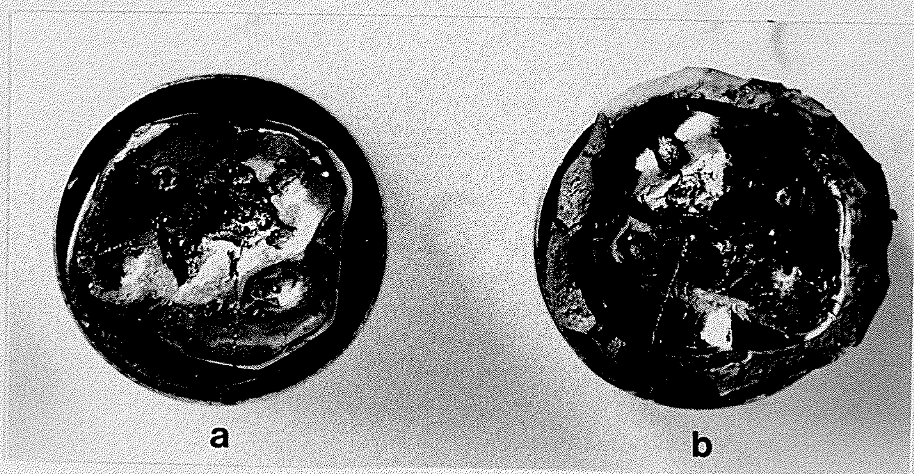
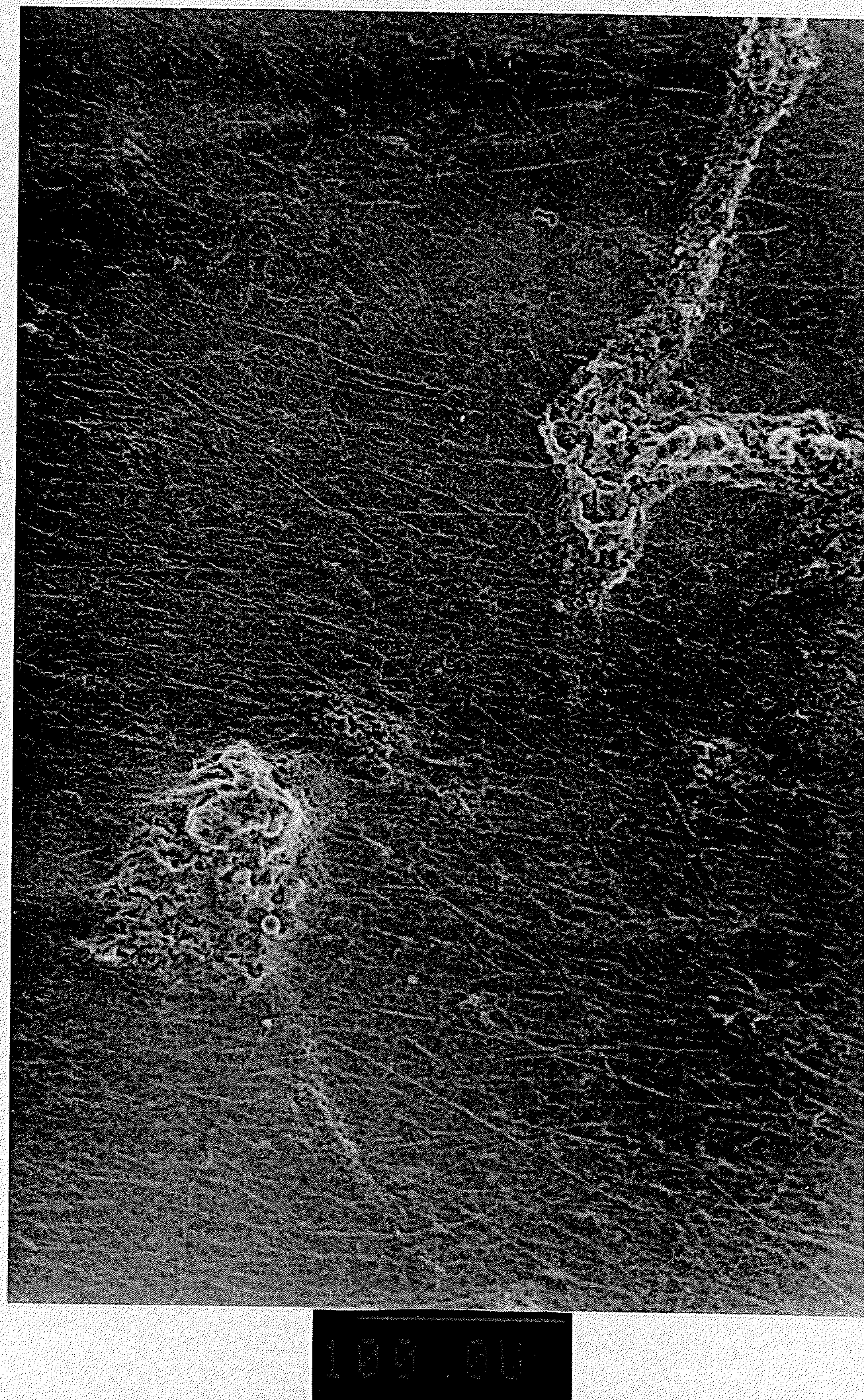


Figure 10. SEM photomicrograph of craze lines on gold-palladium coated impression.

SEM photomicrograph taken at 200X magnification of the negative impression of the tooth surface made with Reprosil polyvinyl siloxane impression material. Thermal contraction of the impression material in the vacuum chamber of the SEM caused crazing of the gold-palladium sputter coating. Surface details were obscured by the craze lines, producing poor images for the identification of microwear features.

Figure 10. Photomicrograph of craze lines on coated impression



4.4 Casting technique

Casting material is required to have many of the same properties as the impression material; that is it must produce high resolution duplicates, have extended dimensional stability at varying temperature and air pressure, and must be compatible with the impression material. At least three different types of materials have been used successfully as casting material in the past, including epimine resin, epoxy cement, and epoxy resin (Barnes 1978; Pameijer and Stallard 1972). Epimine resin in the form of Scutan, a crown and bridge temporary restorative material, is not compatible with polyether rubber, and does not flow as well as some epoxies. Epoxy cement, such as Britfix, is a commercially available household adhesive; as such it is inexpensive and easy to use. The draw back is that it does not have as good microscopic resolution, although some have found it to be adequate for low magnification work. Epoxy embedding material appears to be the material of choice, and is the one that most if not all researchers in the field use at present. As an embedding material, it is crucial for it to be of low viscosity, and be able to penetrate biological tissue completely. For this to happen, epoxies take a long time to set, allowing for thorough flow and penetration of crevices to occur. The draw back is that it takes a long time to set, and often requires regulated and elevated temperatures for optimal curing.

There are many different brands of epoxy embedding materials with different viscosity, hardness, and rates of curing. In this study, Epon epoxy resin (E.F. Fullam) was chosen for its low viscosity and surface hardness after curing.

Epon is a four part liquid system. When mixed together, it has very low viscosity, and will take 48 hours to set. Manufacturer recommendation suggests curing should take place at a temperature of 50-60 degrees celsius for 48 hours. This method was modified to include initial curing at room temperature for 24 hours, followed by 48 hours of curing in a 50 degree curing oven. The reason for modifying the method is to reduce the chance of pitting artifacts that can form during castings; details will be discussed in the following section.

4.5 Replication artifacts

Producing casts for high resolution microscopic studies require a precise technique. Once the proper materials are selected, one must be aware of the limitations of the materials; the handling of the materials will make a drastic difference in the success or failure of the final product.

When making the primary impression, the tooth surface must be clean and dry, as described before, being cognizant that any debris left on the tooth will be replicated as well. Silicone impression materials are hydrophobic, and will not penetrate and replicate any surface covered with moisture. The silicone impression material must be mixed according to manufacturer's recommendations, under controlled temperature and humidity. Setting time must be closely monitored, so as to avoid movement of the specimen or the impression material during the period of setting. The silicone was applied to the surface of the specimen through the tip of a syringe in an even, continuous fashion, to avoid trapping air between the impression and the specimen surface. Once an initial layer of impression material was applied, the surface of interest with the impression material was set into an additional pool of freshly mixed impression material, and left untouched until the impression had fully set. The weight on the tooth will provide pressure to keep the tooth surface in intimate contact to the impression material during the setting period. Any air trapped between the tooth surface and the impression material will show up as positive bubbles on the surface of the casts. Often they were easily recognizable, but depending on the surface of interest, they may obscure the features that were crucial to the study.

Initial curing of the impression material, taking roughly six minutes from the start of mixing, will see approximately 75% of polymerization take place. The remaining polymerization process may continue for days or even weeks after initial setting. As mentioned before, condensation-cure type silicones will eliminate ethanol as a byproduct of polymerization; the addition-cure type, though does not give off byproducts, but may give off unbounded or excess molecular hydrogen as long as polymerization continues. Therefore, any degassing that occurs during the time that uncured epoxy resin is in contact with the impression material will leave trapped air bubbles on the surface of the final casting, resulting in a pitted surface (Gordon 1984d).

To alleviate some of the delayed degassing problems, the manufacturer of Reprosil recommends a delay of 24 hours before pouring the casts, in order to allow for more molecular hydrogen to bond and the excess to escape; while other manufacturers suggest immediate pouring of impressions to minimize polymerization shrinkage. Gordon (1984d) further suggests overcuring of the empty impressions by placing them in a heated environment of 50 degrees celsius for up to 60 minutes, to drive out excess unbonded hydrogen. Although this process may speed the rate of polymerization, it did not seem to be any better a solution than allowing for polymerization to take place naturally. In fact, the added shock of heating and cooling of the impression material may further decrease dimensional stability of the impression, or cause thermal breakdown of the impression material if conditions are not well controlled.

During my trial experiments, degassing artifacts were not a significant problem, when manufacturer's recommendations were followed. In one incident, the curing oven's temperature gauge broke, and the oven temperature was much higher than expected. Significant degassing artifacts were found on almost every cast (Fig. 11). Dimensional changes cannot be measured, but many casts showed signs of warping. The distorted casts had surfaces that were dimensionally distorted, and measurements taken will not be accurate compared to the original; consequently, the impressions were discarded and new ones retaken. One method that was adopted since that time was to allow for 24 hours of epoxy curing to occur at room temperature before placing them in a 50 degree celsius oven. The initial curing at room temperature allows for epoxy to cure to a certain extent without the occurrence of accelerated degassing. Degassing artifacts were not quantitatively compared between different methods, but the latter method produced satisfactory results with no degassing artifacts found on any of the wear facets of interest.

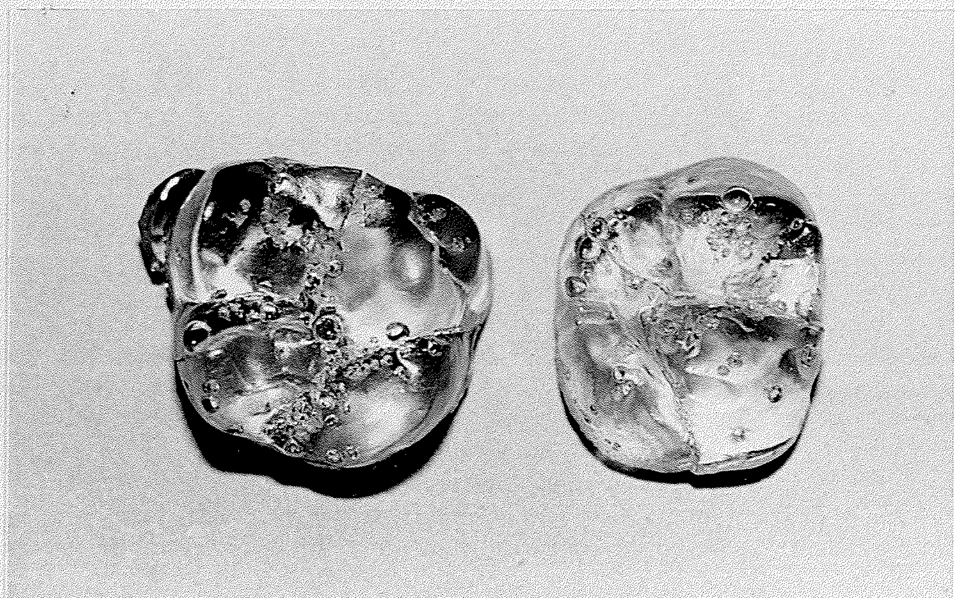
4.6 Preparation of casts for SEM viewing

The casts made from epoxy resin were mounted on half-inch specimen stubs. The bases of the casts were trimmed to be parallel with the occlusal surface of the tooth. The thickness of the bases were also reduced to be as thin as possible, so that the occlusal

Figure 11. Casts of teeth from a Ganj Dareh sample showing degassing artifacts.

Epon casts with degassing artifacts due to gas release from the impression material during curing of Epon epoxy casts. Degassing occurs occasionally due to the continued curing of the impression material over a period of several days, after initial mixing of the impression material.

Figure 11. Casts with degassing artifacts



surface of the cast would be as close to the surface of the specimen stub as possible. By doing so, we were certain that when examined with the SEM, the facets of interest would consistently be at the same distance and orientation from the source of the electron beam. The casts were glued onto the stubs with silver conducting paint, which allowed for proper conductivity to an otherwise non-conductive epoxy cast. The mounted casts are then coated with gold-palladium to provide proper conductivity and reduce surface charging.

Plating of specimens took place in the Hummer V sputter coating machine. A current of 10 mAmps was passed for 2 minutes, providing an 100 angstrom thick coat of gold-palladium on the specimens.

4.7 Scanning electron microscopy of casts

To obtain high quality images of the microscopic wear pattern on the teeth, photomicrographs were taken using scanning electron microscopy. As described in the literature review, early researchers have used both light microscopy for this purpose (Dahlberg and Kinzey 1962; LeJeune and Baron 1973) as well as scanning electron microscopy (Boyde 1967, 1971, 1979, 1981). Scanning electron microscopy has the advantage of providing extremely high magnification and resolution and electronically adjusting contrast; making the images clear and sharp, and easier to interpret. Most importantly, photomicrograph images from SEM have a significantly greater depth of focus than light microscopy, allowing for clarity of features having different depths of the field to be taken on one photomicrograph.

Figure 12. Effect of tilt on photograph contrast.

SEM photomicrographs were taken at 480X magnification of tooth 47 (lower right second molar) from individual 497. Contrast of microwear features against background was examined. Differences in feature contrast were found between photomicrographs taken with and without tilting of the specimen. Increase in feature contrast was found when the specimen was tilted towards the secondary electron collector in the SEM chamber. The increase in contrast between microwear features and its background was created due to the increased shadowing of the depth of the microwear features when the specimen was tilted.

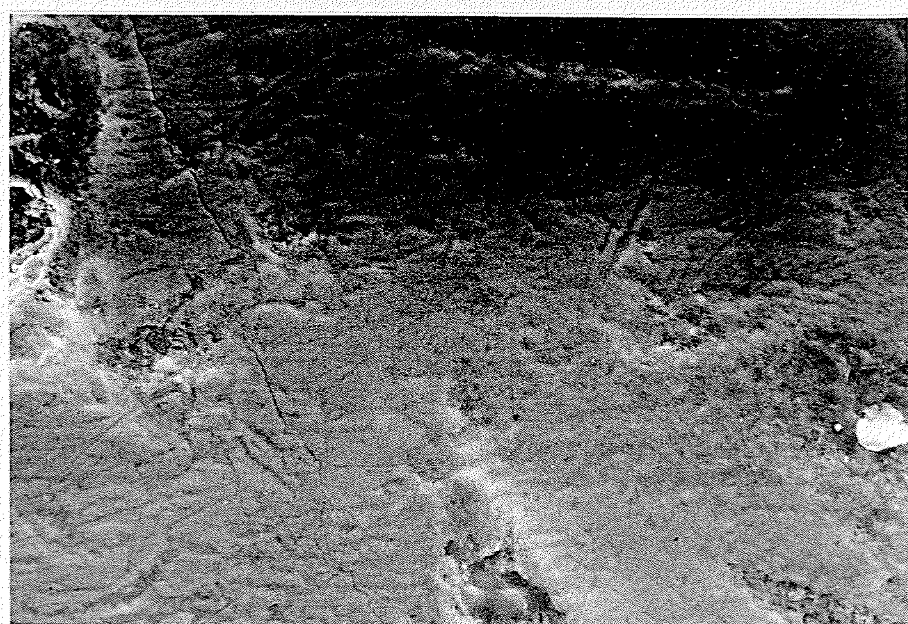
Figure 12a. 10° tilting of the specimen towards the secondary electron collector of the SEM. Notice the increase in contrast of the microwear features found on this photomicrograph when compared with Figure 12b.

Figure 12b. No tilting of the specimen was used in taking of this photomicrograph.

Figure 12. Effect of tilting specimens on photograph contrast

a. 10° Tilt

10.0U



b. No Tilt

10.0U

The casts were examined with a Joel JSM-35C scanning electron microscope with a focal distance of 15mm. Secondary electron images were produced with the electron beam operating at 15KeV accelerating voltage. The specimens were oriented in the specimen chamber with the buccal surface of the tooth at the top when viewed on the CRT. The facets of interest, in this case phase II facets (see facet number 9 in Figure 6), were located by a general survey of the occlusal surface of the tooth. Once found, magnification was increased until the microwear features could be seen on these facets. The numbering system for the facets used was adopted from Kay (1977) with the modification made by Gordon (1982). The work done by these researchers has made detailed distinction and identification of wear facets along with their function possible. In this study, facet number 9 was chosen to represent phase II chewing facets whenever possible, and other phase II facets were used in cases where facet number 9 was not available; due to severe, abnormal, or lack of wear (Fig 6). Approximately 90% of the samples used had identifiable facet 9. An area on the facet with microwear feature patterns and not obscured by artifacts, was selected, and photomicrographs were taken at 480X magnification. Photomicrographs from four areas of the facet were taken, to represent random sampling of that facet. The average microwear pattern found on these four samplings was then used to represent that tooth. Photomicrographs taken at 480X magnification represent approximately $0.033mm^2$ of facet area. The total area sampled by the use of four photomicrographs was approximately $0.132mm^2$. Since the facet itself varied greatly in size, depending on the amount of wear the tooth had experienced, the total sample area represented 5 to 50% of the facet. Photomicrographs were taken with Kodak TMAX 120 film, and subsequently processed with TMAX processing solution.

Standardized orientation of the tooth was established before the photomicrographs was taken. The buccal surface of the teeth were located and placed at the top when seen on the viewing screen. Since the orientation of the microwear features in relation to the electron beam and the collector may affect how well the features were seen (Gordon 1988), it was important to be consistent in orientating the facet of interest. The standardization of orientation will eliminate some of the effects of instrumentation on microwear features perceived.

Early in the preliminary studies, photomicrographs were taken and found to lack significant contrast. Communications with other researchers in the field (Dr. Teaford, Dr. Ungar, personal communications) revealed that a certain amount of tilt is needed to enhance the contrast of the features; some researchers used as high as 45 degrees of tilt, towards the collector. Similarly, a certain amount of tilt was introduced in the present samples, the amount of tilt was dictated by the contrast of the surface features (Fig 12). The tilt of the sample was always towards the collector, that being around the buccal-lingual axis of the sample, when viewed on the CRT. The amount of tilt was recorded and compensated for in the analysis of the photomicrograph, as described in the next section.

4.8 Image analysis

The analysis of the image consists of quantifying the microwear features. Parameters measured consisted of; the number of features, the size of the features, the direction of feature as they were formed, and the total area of facet surface that appears on the photomicrograph. Traditionally, all these measurements are done manually; each feature on the photomicrograph was identified, and their length, width, and direction were measured and recorded. This has proved to be an extremely tedious process, both due to the number of features that were present, as well as the way that features overlapped one another and were possibly interspersed with feature artifacts. One of the goals of this project was to develop a computer image analysis program to automate the data collection. With help from Dr. Stephen Simons of the Grain Commission, and Dr. Salah Hathout of the University of Winnipeg, different ways of achieving automated data collection were attempted. Unfortunately, we were only able to achieve a semi-automated process.

Dr. Simons modified the IBAS image analysis program to provide a semi-automated image processing system for our purposes. Negative film was placed on an illuminated table. The photographed image was then picked up by a Nikon camera equipped with a Nikkor Micro 105mm lens. The original orientation of the tooth was preserved by orientating the negative consistently the same way; the digitized image would then be in the same orientation as the image seen originally with the SEM. Kontron's IBAS1 20.669 386DX computer ran the DOS managed IBAS (Release 2.0) image processing software, which imported the digitized image. Once the image had been imported, it was displayed on a high resolution (1024x1024 pixels) Sony monitor, making the displayed image roughly two times larger than the original photomicrograph.

Since the photomicrographs were taken with the specimen tilted towards the collector, occurring around the Y-axis of the image, the image was foreshortened in the X-axis dimension. To compensate for the foreshortening, the computer stretched the image along the X-axis only, according to the amount of tilt. The proportion of foreshortening was equivalent to the cosine of the angle of tilt. Since the angle of tilt was originally recorded when the photomicrographs were taken, the angle can be entered into the image analysis system, and the image dimension along the X-axis was then calculated and compensated accordingly.

Image manipulation was possible with the various enhancement programs that the IBAS image analysis program has in its software. For our purposes, the contrast of the image can be enhanced to produce optimal feature identification. Since the black and white image was produced according to 255 different shades of grey, we were able to set upper and lower limits of the shades of grey that the features of interest are occupying, with the program then stretching that range to include all 255 shades of grey, effectively increasing the contrast of the features of interest.

Next, the program was provided with proper scaling measurements, by indicating the length of the scaling bar with the cursor. Once the information of scale was entered, all subsequent lengths and widths identified with the use of the cursor by the operator will be calculated to scale by the program. Since the image displayed may include areas that were unwanted or made undesirable by artifacts, the program prompted the operator to identify the area of interest with the cursor. The total area of interest thus identified was then calculated and recorded by the program to give the true area measurement where the microwear features were located, excluding areas where undesirable artifacts may be present. The program then prompted the operator to enter the long axis or length of a feature by placing the cursor first at the formation end of the feature, and then at the tail end of the feature, identifying the greatest diameter of the feature. The short axis or width of the feature was then identified by the operator as being the widest point on the feature that was perpendicular to the long axis of the feature, and entered in to the program with the use of the cursor. The line drawn between the two ends identifying the length of the feature was calculated on an angular scale to identify the direction of the feature; with zero degrees at the 3 o'clock position, working around to 359 degrees in a counter-clockwise rotation. All the feature parameters were identified by the operator. An example of microwear feature identification on a photomicrograph is illustrated in Figure 13. The routine was repeated until all the features were located on a given field on the photomicrograph. The information regarding number, length, width, direction of features, and total area of tooth surface where the features were located, was stored in a computer file. The file was named alpha-numerically, identifying which individual the tooth was from, the tooth number, which replica of that tooth, and which facet on that tooth.

4.9 Data manipulation

By using the data collected the following were calculated for each feature:

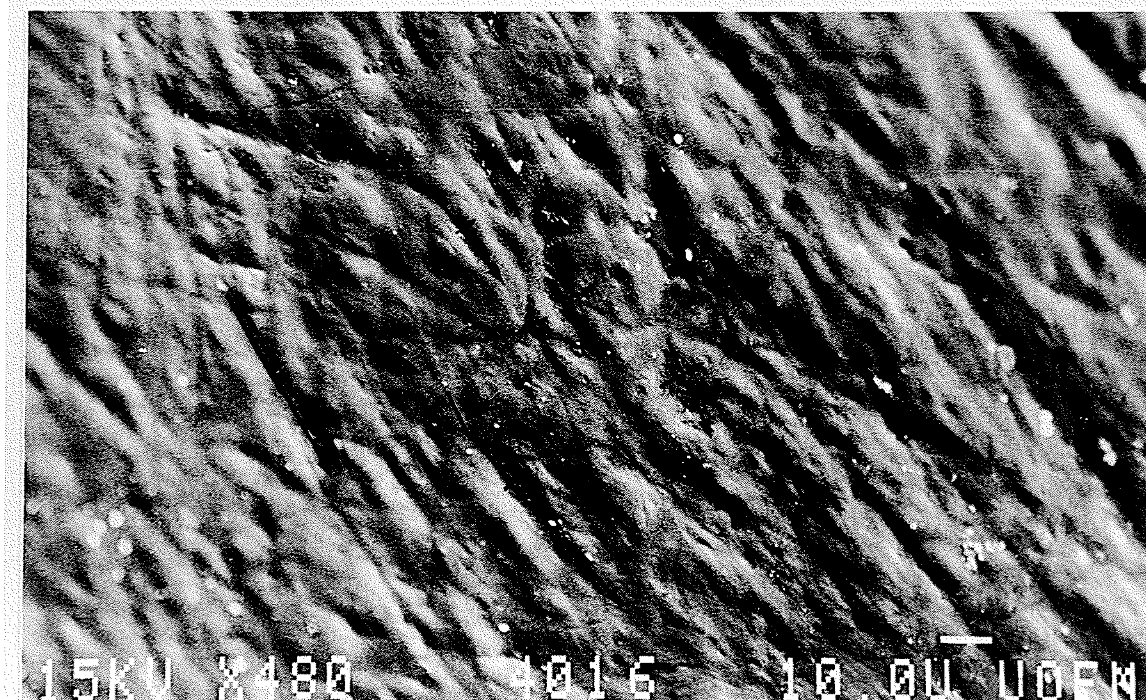
1) Pit: Any feature with long axis to short axis ratio of less than or equal to 4.

2) Scratch: Any feature with long axis to short axis ratio of greater than 4.

Figure 13. Method of measuring microwear feature dimensions.

SEM photomicrograph taken at 480X magnification of tooth 16 (upper right first molar) from individual 40 of the Ganj Dareh samples. The image was digitized and shown on a high resolution computer monitor. Microwear features were identified by the operator. The long axis or length of feature was identified by placing the computer cursor first at the formation end of the feature, and then at the tail end of the feature, identifying the greatest diameter of the feature. The short axis or width of the feature was then identified by the operator at the widest point on the feature that was perpendicular to the long axis of the feature, and entered into the program with the use of the cursor. The line drawn between the two ends identified as the length of the feature was calculated on an angular scale to identify the direction of the feature; with zero degrees at 3 o'clock position, working around to 359 degrees in a counter-clockwise rotation. Examples of features with their length, width and orientation identified are shown by the arrows on the photomicrograph.

Figure 13. Method of measuring microwear feature dimensions.



3) Feature Area: Estimated by using an ellipsoid formula; that is multiplying feature long axis radius by feature short axis radius, by 3.14.

Analysis of microwear patterns has traditionally concentrated on pit to scratch ratio for a given field. This method takes into account only the number of pits or scratches, and does not take into account the size of features. Visual examination of the microwear features usually includes not only how many features there are, but also how big or small the features are. Large pits and scratches, although fewer in number, will indicate a highly destructive diet. Therefore, if only the number of features were considered, one would reach an erroneous conclusion regarding the abrasiveness of the diet. Similarly fine scratches and small pits, not just their numbers, will indicate a less abrasive diet. This problem was addressed by comparing both the size and the number of feature, instead of just the number of features. Therefore, the following parameters were calculated for each micrograph in this study.

Traditional Parameters:

1) Feature density (FD): number of features per 1,000 μm^2.

2) Pit density (PD): number of pits per 1,000 μm^2.

3) Scratch density (SD): number of scratches per 1,000 μm^2.

4) Pit to Scratch ratio (P/S): number of pits divided by number of scratches in a given field.

New Parameters:

5) Average Length of Feature (A_Length): the average length of the features.

6) Average width of feature (A_Width): the average width of the features.

7) Average Ratio (A_Ratio): average feature length divided by average feature width. This gives the average length to width ratio that will describe the shape of an average feature found in the field, with the assumption that pits and scratches are part of a single microwear continuum.

8) Proportional Area Devoted to Features (FA): sum of all feature areas divided by the field area.

9) Proportional Area Devoted to Pits (PA): sum of all pit areas divided by the field area.

10) Proportional Area Devoted to Scratches (SA): sum of all scratch areas divided by the field area.

11) Pit-Area to Scratch-Area Ratio (PA/SA): pit-area divided by scratch- area in a given field. It is the area equivalent of pit to scratch ratio.

Microwear features' data from a total of 92 sample areas were collected, representing four sample areas from each of 23 teeth used in this study. The 23 teeth were collected from 9 different individuals with molars that were suitable for microwear analysis (see Table 1 for detailed information regarding each individual). Data from the microwear features were then used to calculate the values of each of the above eleven parameters. The data for each of the parameters were then imported into The University of Manitoba's main frame computer. The SAS program was employed for statistical analyses of the significant parameters identified for this study.

4.10 Statistical analyses

Traditionally, quantitative microwear studies have compared pit to scratch ratio, pit density, and total feature density between different groups to infer abrasiveness of diets. As indicated in the literature, the separation of features into categories appears to be somewhat arbitrary. To test if pits and scratches are simply features on opposite poles of a microwear continuum, the features found in each photomicrograph have been plotted with respect to their length to width ratio in 1 unit increments. The resulting distribution of feature length to width ratio plots were examined for clustering of ratios around different means. A distribution plot of two or more distinct entities on one graph will exhibit signs of two or more peaks ratios in the plot, indicating the overlapping of two or more bell-shaped normal distributions with separate means and variances.

As mentioned previously, a new method of microwear analysis involving the area of features was being tested in this study to determine if it will predict the abrasiveness of diet in different individuals. Pearson correlation coefficient analysis was conducted to determine the correlation between the parameters involving feature area and parameters using number of features. A high correlation of feature area parameters to feature number parameters would suggest little or no difference in the results when either one was used in microwear analysis. The correlation test is intended to detect correlation of data from two samples. The real sample units in this study are the four sample areas taken from different regions of each tooth. The basic unit for subsequent comparisons for this study was the tooth itself, therefore, the data from all the sample areas originating from the same tooth were pooled, and the correlation of parameters was done with the tooth as the basic unit. Since the correlation test was not intended for use with pooled data, the results from these correlation tests need to be viewed with caution. The results from this test were intended to give a general relationship of the parameters, and were not used in subsequent statistical testing.

The comparisons made for between and within individual differences in this study were tested by using one or two way analysis of variance models. The significance level of the F values are presented in tabular form. The critical p value is normally accepted at 0.05, but due to the large numbers of comparisons, the critical p value for all the comparisons was set at 0.01. Some may argue that the critical value should be more stringent by dividing 0.05 by the number of comparisons made (Bonferroni correction) to reduce type I error; but all the parameters used in this study have been chosen for their potential ability to discriminate microwear pattern, and the use of the Bonferroni correction would be too conservative and could introduce excessive type II error (Hassard 1991).

Those individuals with more than one tooth available, were tested for within individual differences. By using a two way analysis of variance, involving the interaction of seven different individuals and four different positions of teeth, it was possible to compare the top teeth (teeth number 16 and 26) to bottom teeth (teeth number 36 and 46); and compare teeth on the right side (16 and 46) to their complimentary on the left side (36 and 46) for within individual differences with respect to all the parameters established above. In the case of top to bottom comparison, tooth 16 was paired with 46, and 26 was paired with 36 in each individual. The side to side comparison was conducted with tooth 16 paired with 26, and 36 paired with 46 in each individual (refer to Table 1 for detailed tooth and individuals available for comparisons). To avoid *a priori* assumption of insignificant differences between right and left teeth, the two top teeth were not pooled for the comparison to the two lower teeth. Same reasoning was adopted for right to left comparisons. Examination of the literature revealed that microwear pattern on the first molar, regardless of which quadrant of the mouth it came from, has always been assumed to be the same within an individual. This assumption neglects consideration of the possibility of dominant side function, or habits that occur only in one quadrant of teeth. The tests for within individual differences in this sample will allow for detection of such occurrences. In the event that no significance was found for any of the comparisons tested, then all the teeth within an individual can be pooled for later between individual testing.

Testing for between individual differences of the same parameters as described above was also done. One way analysis of variance was used for these comparisons, with data from all the teeth within an individual pooled together if there was no significant within individual differences found; and the same tooth from all the individuals were compared separately in cases where within individual tooth position proved to be significant.

Sex and age differences between individuals were also examined, for those individuals that had their sex and age identified. Two-way analysis of variance was used to detect sex differences and age differences. For the age analysis, individuals were group according to child (individuals 12 years old or under) and adults (individuals over the age of 12). There were a total of two individual 12 years old or under, and 7 individuals over 12 years of age. Sex differences were examined in 2 females and 5 males. Two of the nine individuals could not sexed due to a lack of physical characteristics available for definitive sex discrimination.

The individuals within this group were compared with the known microwear data of several primate species with different diets, derived from an earlier study by Teaford and Walker (1984). The parameters they chose for diet discrimination were: average feature length, average feature width, and pit to scratch ratio. Since the pit to scratch ratio they had chosen was 10, the pit to scratch data used here have been converted, in this section only, to be directly comparable to their results.

The only available prehistoric human data that were contemporary to the present study were published by Molleson *et al.* (1993). She used proportional pit area and feature density for diet discrimination between groups. Data from the present study were converted for direct comparison. One major criticism for this comparison of data is that Molleson has conducted her microwear analysis on photomicrographs taken at 180X magnification. As mentioned in the literature review, results obtained from analyses of microwear features at different magnifications may not be directly comparable; any comparisons made in this fashion should be regarded with caution.

4.11 Modern samples

The modern sample in this study involved two young adults, Subject A was a 23 year old male, and Subject B was a 21 year old female. Both subjects were recruited from the student population at The Faculty of Dentistry, University of Manitoba. Each of the subjects had a thorough dental examination before the start of the study, including a clinical exam and questionnaire as to their functional and parafunctional activities. No abnormal tooth positioning or wear were found on any of the teeth in the two subjects. During the functional examinations, however, Subject A confessed to a clenching habit that occurred during the times of stress; no significant wear facets were found to suggest excessive wear due to this parafunction. Subject B confessed to general muscle and temporomandibular joint pain occurring occasionally over the past 6 or more years. She reported no changes in her diet pattern due to this condition, with the exception of her discontinuation of gum chewing.

Impressions of the two first molars on their dominant side (which they had identified as the side they most often chew with) were taken on two separate occasions, eight days apart. Casts were made of these teeth with the same material and method as for the prehistoric human samples, and representative SEM photomicrographs were taken of the occlusal facets. Evaluations of their microwear patterns were done on a qualitative basis. Microwear features on the first of the two successive photomicrographs were identified and counted. New features not found on the first photomicrograph were identified and counted on the final photomicrograph. The number of new features formed over the eight days would give an estimated rate of turnover of microwear features found in that facet. The rate of turnover in microwear features will help us determine the amount of tooth wear that individual was experiencing. The possible causes of tooth wear will be explored in the discussion section.

5.0 RESULTS

5.1 Ganj Dareh material

5.1.1. Microwear Analysis on a Continuous Scale: Test for Validity

When the distribution of length to width ratio of the features found on each tooth was plotted in whole number increments, the resulting graphs had the appearance of being continuous in nature (Fig. 14).[3] There was only one peak in frequency, and the distribution was decidedly skewed to the left. The mean and standard deviation of the feature distribution are listed with the distribution graphs found in Figure 14.

5.1.2. Correlation of Parameters Used in Microwear Analysis

Correlation analyses between parameters that utilize both the number and area of features were conducted with the Pearson correlation coefficient analyses. The results are presented in Table 2.[4]

Correlation between pit to scratch ratio (P/S) and pit-area to scratch-area ratio (PA/SA) was not very strong; an overall correlation of 0.67 was found. Since pit to scratch ratio is used most often for microwear analyses and dietary discrimination, it was important to establish a positive if not a strong correlation between the new parameter pit-area to scratch-area ratio.

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[3] All figures referred in the Results section are located at the end of the section.

[4] All tables referred in the Results section are located at the end of the section.

Average length-to-width ratio (A_Ratio) is the measure used in continuous methods of microwear analysis. There was significant negative correlation between A_Ratio and pit-to-scratch ratio (-0.68 for all the teeth combined). Average Ratio was less negatively correlated with pit-area to scratch-area ratio (-0.56 for all the teeth combined). This was somewhat expected, and reflects the fact that PA/SA, unlike A_Ratio, took into account the size of each feature.

Feature density (FD) correlated reasonably with both pit density (PD) and scratch density (SD) (0.65 and 0.73 respectively for all the teeth combined). This, when considered together with the fact the P/S shows low correlation with FD, provides evidence to contradict Gordon's observation that scratch density stays fairly constant, and it is the pit density changes that affects the pit to scratch ratio (Gordon 1982).

Proportional area of features (FA) correlated somewhat with proportional area of pit (PA) and proportional area of scratches (SA) (0.68 and 0.62 respectively for all the teeth combined). The correlations were not strong, but followed the same pattern as feature density correlations discussed above.

SD and SA showed good correlation (0.73 for all the teeth combined), indicating that scratch size and shape were reasonably uniform on most teeth. PD and SD, on the other hand, showed very low correlation, indicating no relationship of pit size to the number of pits present in the field.

Average feature length (A_ Length) and average feature width (A_Width) both correlate with average ratio (A_Ratio). This was expected since average ratio is derived from A_Length divided by A_Width. What is worth noting is that A_length was much more strongly correlated with A_Ratio; thus demonstrating that A_Ratio was more affected by a change in the length of features. Since A_Length had a weak negative correlation with A_Width, the tendency was for the feature width to stay the same or decrease, as the feature length increased; that is, there was a trend toward long features that were thin, and short features that were wide.

Overall, there appeared to be a weak positive correlation between the parameters that measured the number of features with those that measured the area of features.

5.1.3. Within Individual Differences

Within individual differences explores the possibility of differing microwear patterns between teeth in the upper and lower jaw, as well as differences between teeth on the right side and teeth on the left side. The significance levels of the variance ratio of each of the paired comparisons are found in Table 3.

The only significant differences were found between tooth 16 and 46 with respect to SD and A_Width. When the comparisons at the individual level were examined, the only significant differences for SD and A_Width were found between teeth 16 and 46 (right upper and lower first molars) in individual 20.

5.1.4. Between Individual Differences

Since the results of within individual differences showing no significant result except as just noted, it was decided to pool all the teeth within each individual to test for between individual differences. The significance values of the variance ratios for individual comparisons are listed in Table 4.

Significant differences were found between individuals for the parameters of average ratio, feature density, scratch density, pit density, pit to scratch ratio, and pit-area to scratch-area ratio. No significant differences were found for the parameters of proportional area of pits, proportional area of features, proportional area of scratches. When the individuals were grouped according to children (12 and under), and adults (over 12), no significant differences were found for any of the parameters tested. No significant sex differences were found for any of the parameters tested.

Since statistical analysis cannot be conducted to indicate whether some or all of the individuals were different from one another due to the unbalanced sampling of individuals, relationships of individual microwear pattern were examined by the use of graphic methods. The values of the eleven parameters for each of the nine individuals are listed in Table 5. Significant separation of the nine Individuals by the parameters used are illustrated in the parameter bar graphs showing the individual parameter values (Fig. 15).

The use of two parameters at once for individual separations are illustrated by the use of PA/SA vs. FD, P/S vs. FD, A_Ratio vs. FD, and A_Length vs. A_Width scatter plots (Fig. 16). By considering two parameters at once in separating individuals, we were able to increase the discriminatory power of the microwear analysis. Two of these scatter plots, A_Length vs. A_Width, and A_Ratio vs. FD, when considered together, gave the ability to consider size, shape, and number of features for a comprehensive examination of all characteristics of microwear features. The resulting separation of individuals with this method has the best discriminatory ability. The individuals that showed significant differences by this method are listed in Table 6, and their microwear patterns are illustrated by the representative photomicrographs found in Figure 17. Individual 13 appeared to have a microwear pattern that was predominantly scratches (Fig. 17a). Individuals 16 and 23 (Fig. 17b and 17c respectively) appeared to have a much greater percentage of pits in their microwear pattern than that of individual 13. Individual 40 (Fig. 17d) had a pit to scratch ratio, in his microwear pattern, that was between individual 13 and 16, but there appeared to be a greater number of features on this photomicrograph.

5.1.5. Comparison to Other Studies

The microwear pattern from the present study was compared with seven extant species of primates. Data regarding the feature length and width, feature average length to width ratio, and proportional number of pits are presented in graphic form in Figure 18. The microwear pattern of the Ganj Dareh individuals matched the microwear pattern of those species that were identified as mixed feeders. In Figure 18a, Ganj Dareh individuals were closely matched with *P. troglodytes*, a known mixed feeder of hard and soft diets.

Ganj Dareh data were also compared to those of Mesolithic (1a), Neolithic (2A and 2B) and modern humans from 18th century Spitalfields and Abu Hureyra (2C), found in Molleson's *et al.* (1993) study. The individuals used for these two studies were plotted on a graph of Area devoted to pits vs. total number of features per 0.11 $mm^2/100$ (Fig. 19). The results showed that Ganj Dareh individuals had microwear patterns that most resembled modern human samples. The discrepancy in the methods used between Molleson's study and the present study may prevent valid direct comparison of results.

5.2 Modern samples

Representative photomicrographs of microwear features found on the occlusal surfaces of the first molars of the two subjects in the modern sample are illustrated in Figure 20. Individual A (Fig. 20, a and b) had a significant amount of microwear features on cusp tip facets only, there were no facets in the area of the central groove (facet 9 area), and microwear features were sparse or none in those area. Individual B also had distinct cusp tip facets (Fig. 20, c and d), without any facets in the fossa area; but microwear features were found in the region of where facet 9 should be (Fig. 20e).

Cusp tip facets, in general, showed relatively few features, with the majority of them being large pits or gouges. Microwear pattern in the fossa regions, in individual B, showed relatively few features, tending to be smaller in size. There were relatively more scratches in the fossa regions, with no apparent agreement as to their orientation.

Individual A appeared to have larger, deeper pits than individual B, with parallel sets of very straight scratches that were asymmetric in shape; giving the impression that each set was formed from a single abrasive event. The sets of parallel scratches do not always run in the same direction, and may not be functionally related to the rest of the microwear features.

Individual B also had large pits on the cusp tip facets, but they did not appear to be as deep or as well defined as the pits found in individual A. The microwear features found in the fossa regions were smaller and contained less pits. Enamel prism relief can be clearly seen on two out of the four photomicrographs taken in this region (Fig. 20e and 20f).

Successive impressions were made for the first molars of each of the subjects over a period of 8 days. Initial and final photomicrographs of specific areas on these teeth are illustrated in Figure 21. Very few new features can be found on the final photomicrographs.

For individual A, the photomicrographs of cusp tip facets were compared (Fig. 21a and 21b). On the initial photomicrograph 149 features were found, with 5 new features found on the final photomicrograph. The proportion of new features over 8 days was 0.0336. At this rate, it would take 238 days to replace all the existing features.

For individual B, the photomicrographs of facet 9 region were compared (Fig. 21c and 21d). On the initial photomicrograph, 177 features were found, with 2 new features found on the final photomicrograph of the same region. The proportion of new features over 8 days was .0113. At this rate of wear, it would take 707 days to replace all the existing features.

Figure 14. Distribution of length to width ratio of features.

The distribution of microwear features' length to width ratio was plotted on a graph of length to width ratio vs. number of features, for features found on one tooth from each of the nine individuals from Ganj Dareh samples.

To test if pits and scratches are simply features on opposite poles of a microwear continuum, the features found in each photomicrograph with respect to their length to width ratio was plotted in 1 unit increments. The resulting distribution of feature length to width ratio plots were examined for clustering of ratios around different means. A distribution plot of two or more distinct entities on one graph will exhibit signs of two or more peak ratios in the plot, indicating the overlapping of two or more bell-shaped normal distributions with separate means and variances. The resulting plots presented here appear to have only one peak, which tapered off towards the right. From the visual examination of these graphs, the length to width ratio of features were assumed to be continuous in nature.

Figure 14a. Feature distribution from tooth 16, individual 10.

Figure 14b. Feature distribution from tooth 36, individual 13.

Figure 14c. Feature distribution from tooth 36, individual 13a.

Figure 14d. Feature distribution from tooth 46, individual 15.

Figure 14e. Feature distribution from tooth 46, individual 16.

Figure 14f. Feature distribution from tooth 36, individual 17.

Figure 14g. Feature distribution from tooth 16, individual 20.

Figure 14h. Feature distribution from tooth 46, individual 23.

Figure 14i. Feature distribution from tooth 36, individual 40.

Figure 14. Distribution of length to width ratio of features.

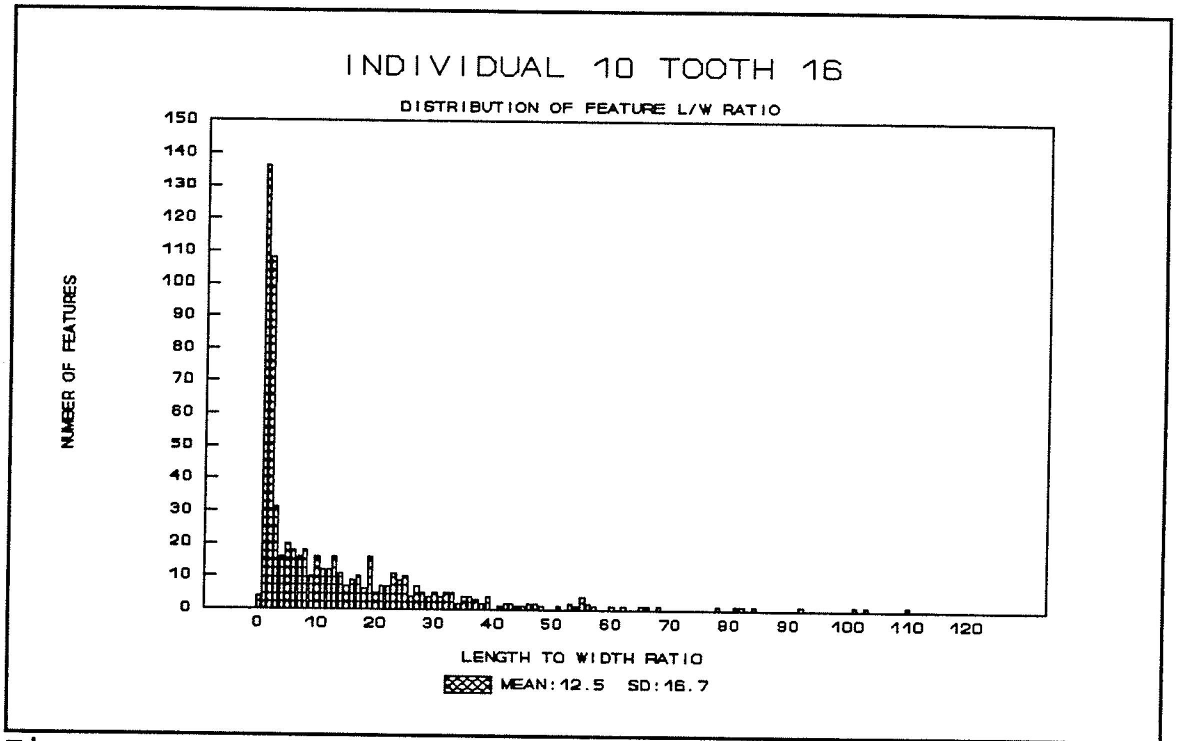


Figure 14a

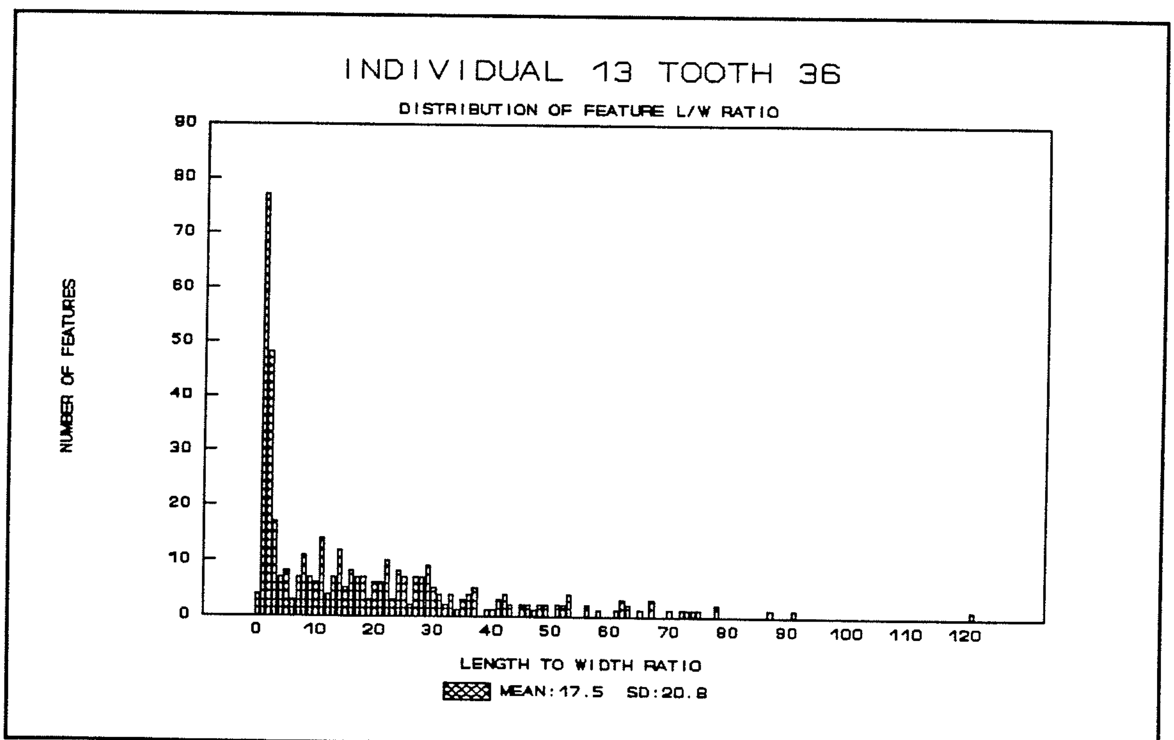


Figure 14b

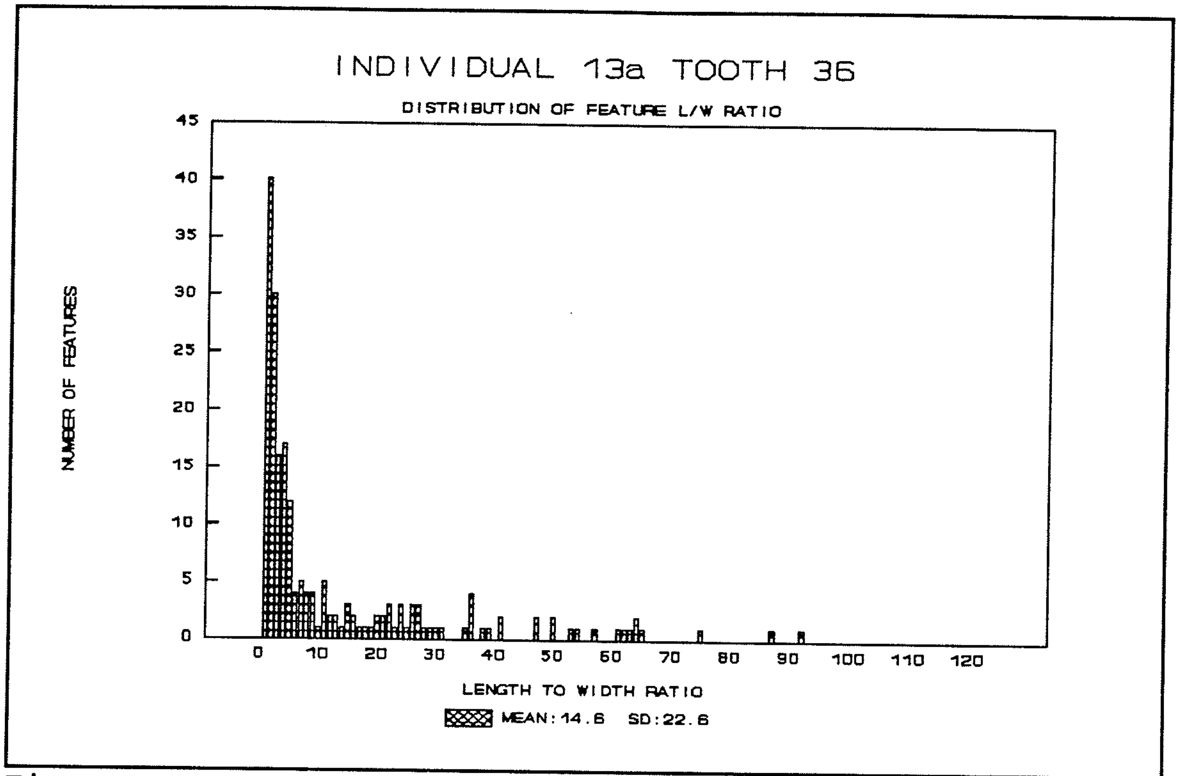


Figure 14c

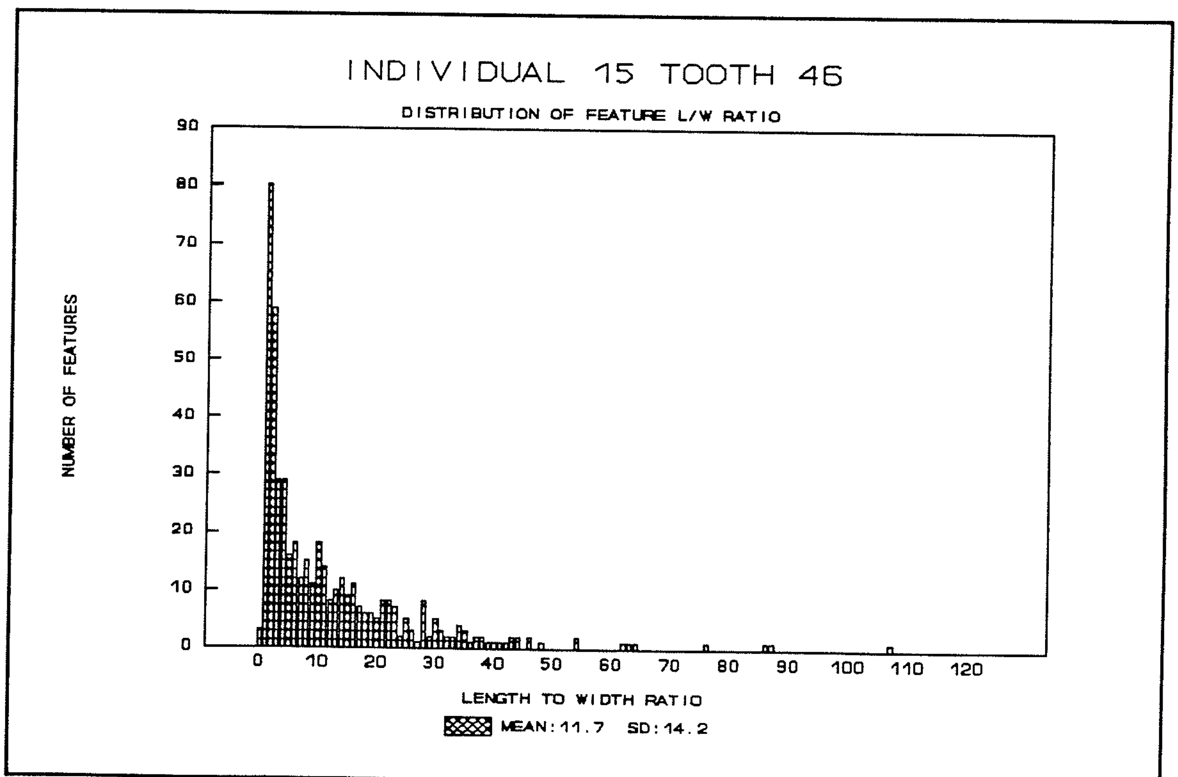


Figure 14d

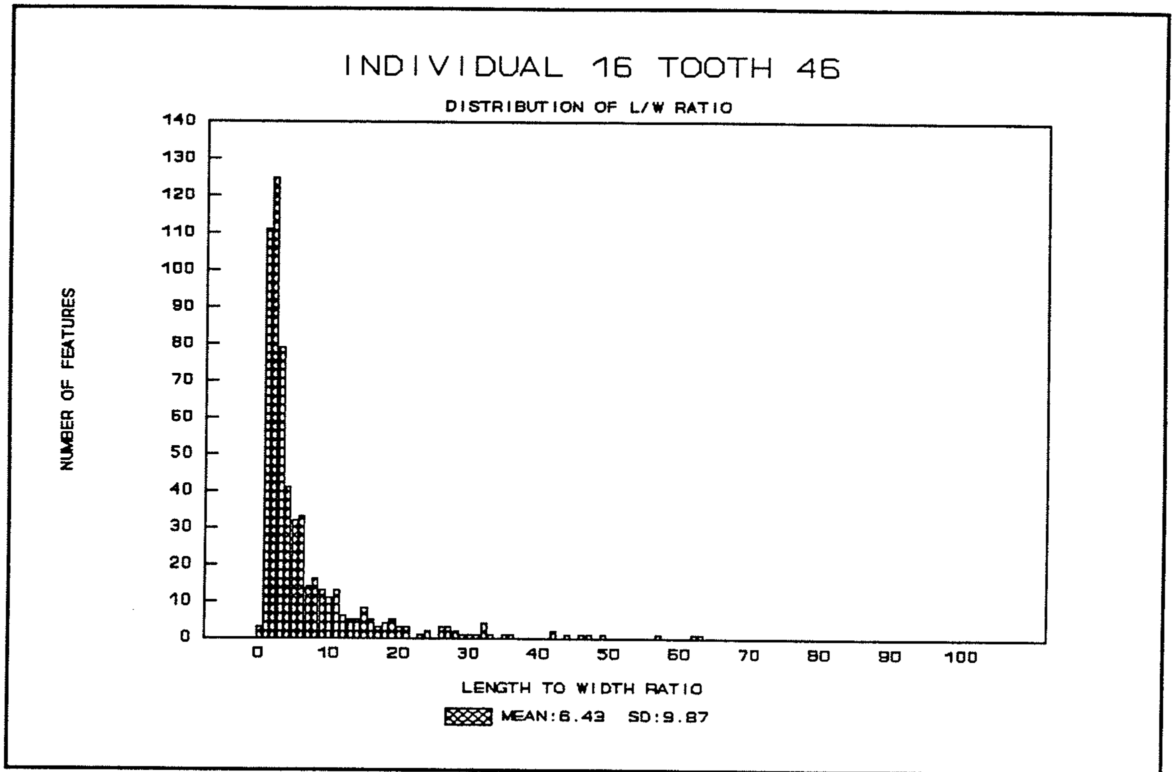


Figure 14e

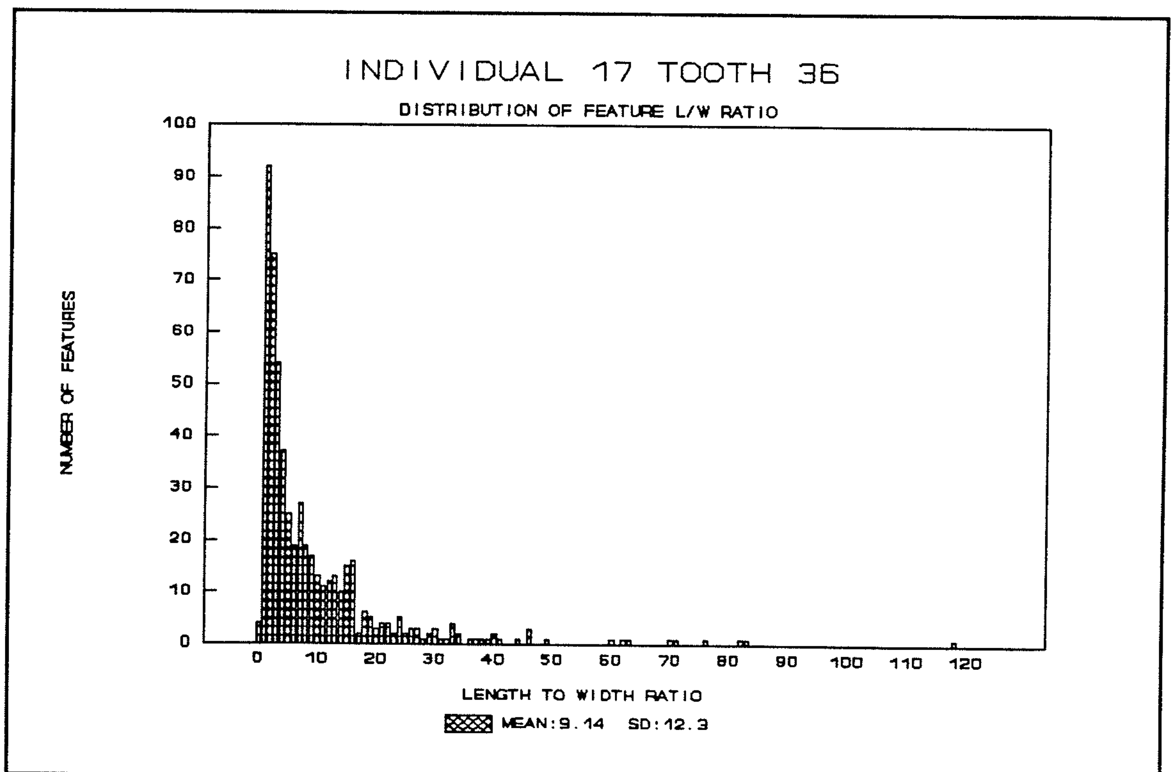


Figure 14f

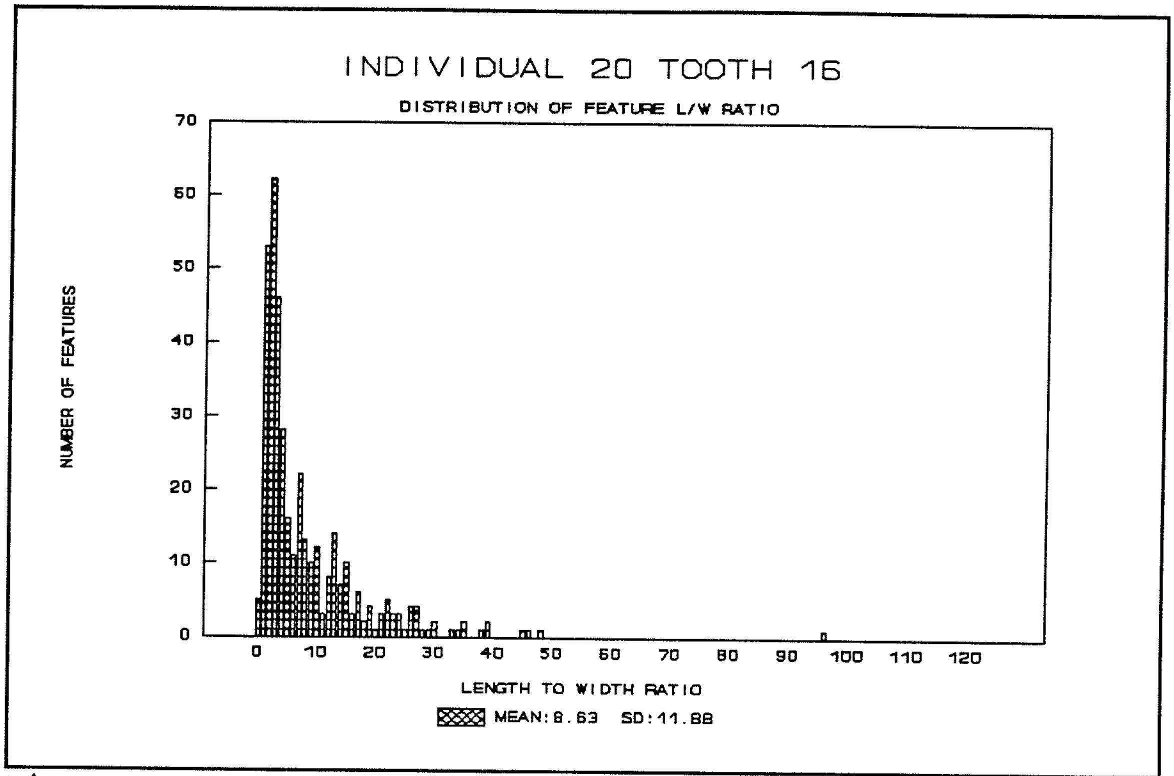


Figure 14g

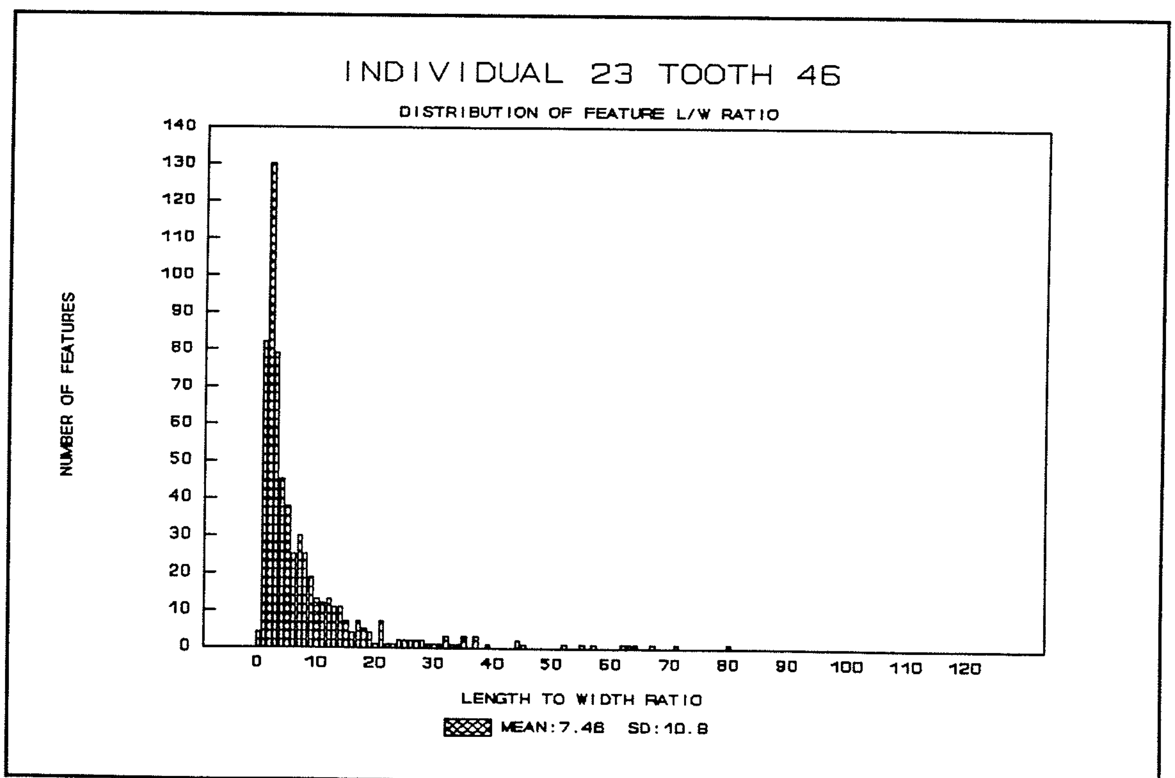


Figure 14h

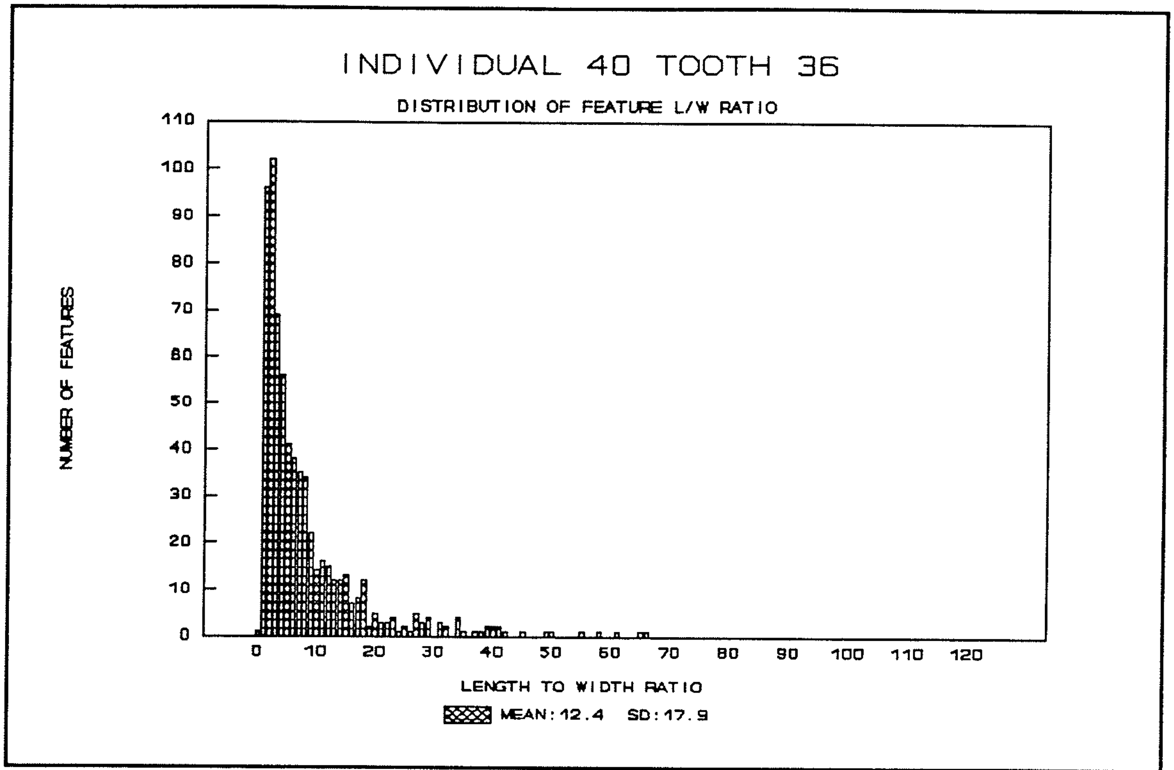

INDIVIDUAL 40 TOOTH 36
DISTRIBUTION OF FEATURE L/W RATIO
NUMBER OF FEATURES
110
100
90
80
70
60
50
40
30
20
10
0
0
10
20
30
40
50
60
70
80
90
100
110
120
LENGTH TO WIDTH RATIO
MEAN: 12.4 SD: 17.9

Figure 14i

Figure 15. Differences between Ganj Dareh individuals according to significant microwear analysis parameters.

Since specific differences between individuals of the Ganj Dareh sample cannot be tested statistically, graphic representation of the parameter values for each individuals were plotted graphically, individual differences that the parameters had detected can be examined visually. All the bar graphs were displayed with the individuals in ranked order according their average length to width ratio.

Figure 15a. Bar graph of individuals with their average length to width ratio values.

Figure 15b. Bar graph of individuals with their average feature length values.

Figure 15c. Bar graph of individuals with their average feature width values.

Figure 15d. Bar graph of individuals with their feature density values.

Figure 15e. Bar graph of individuals with their scratch density values.

Figure 15f. Bar graph of individuals with their pit density values.

Figure 15g. Bar graph of individuals with their pit to scratch ratio values.

Figure 15h. Bar graph of individuals with their pit-area to scratch-area values.

Figure 15. Differences between Ganj Dareh individuals according to significant microwear analysis parameters.

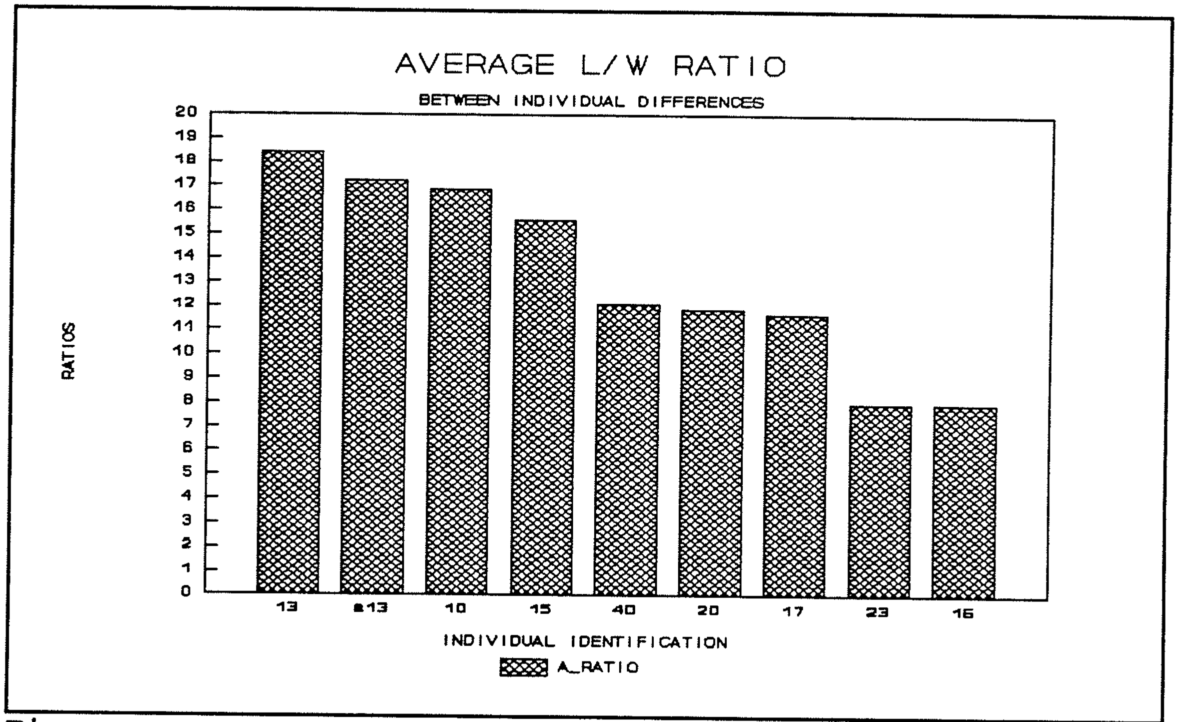


Figure 15a

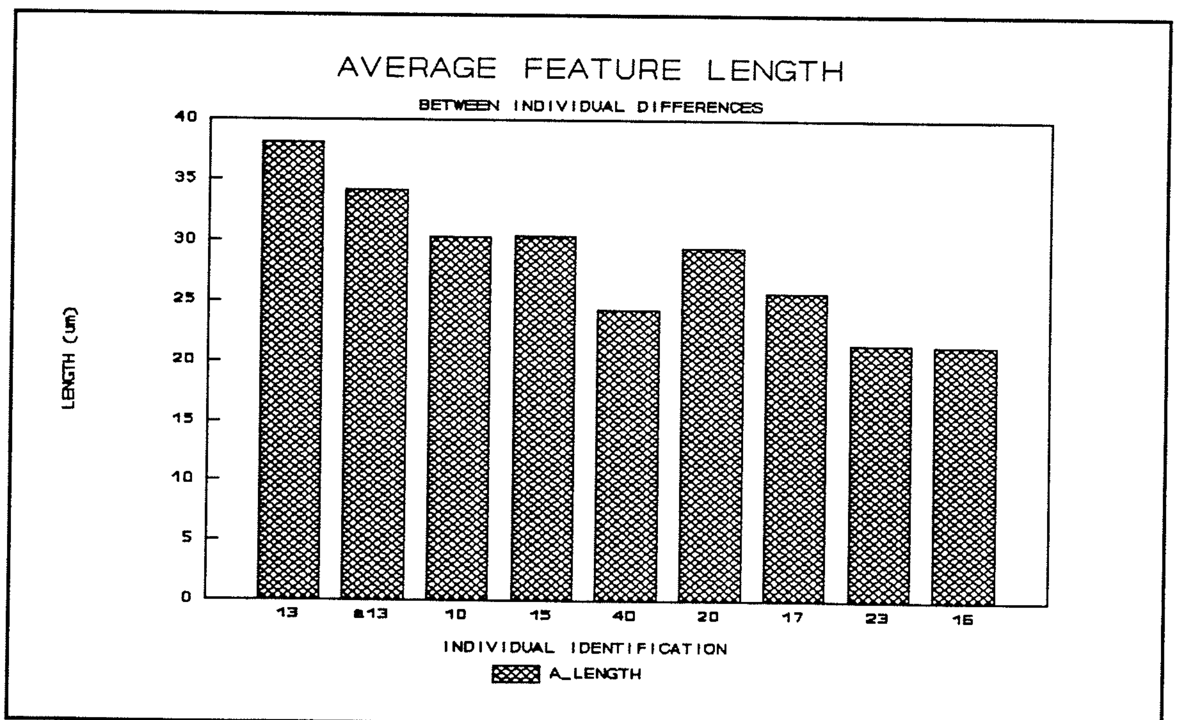


Figure 15b

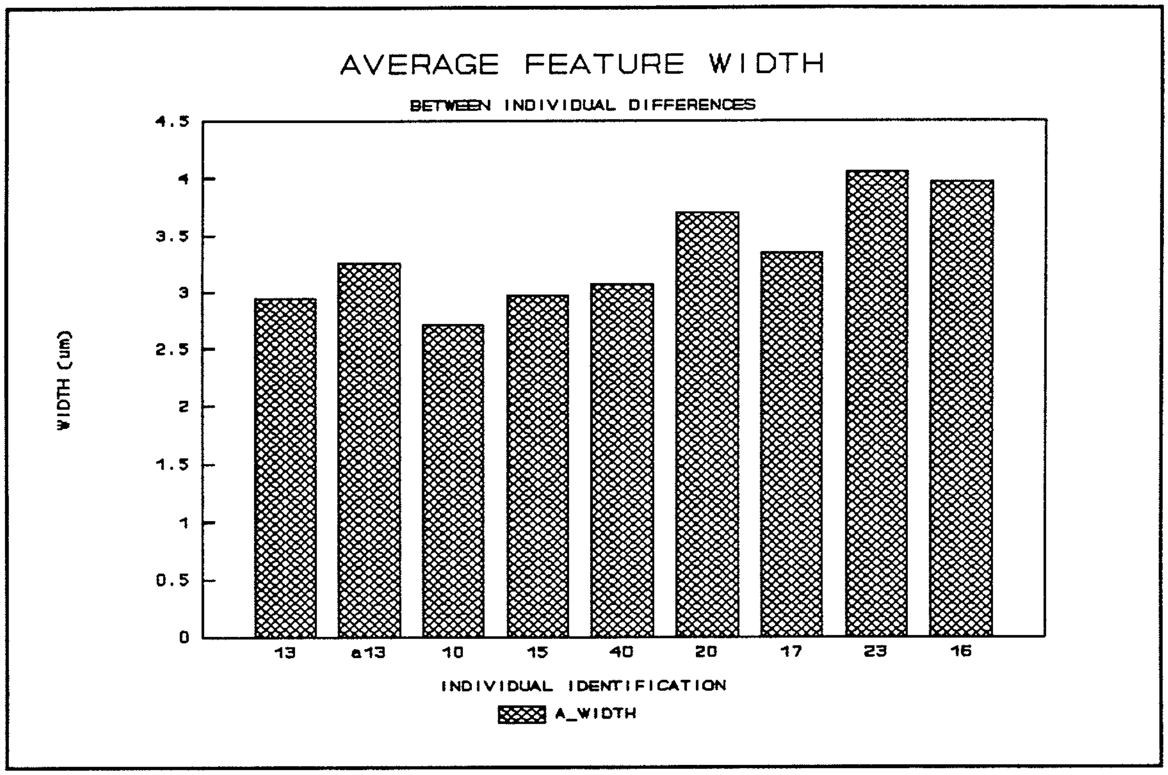


Figure 15c

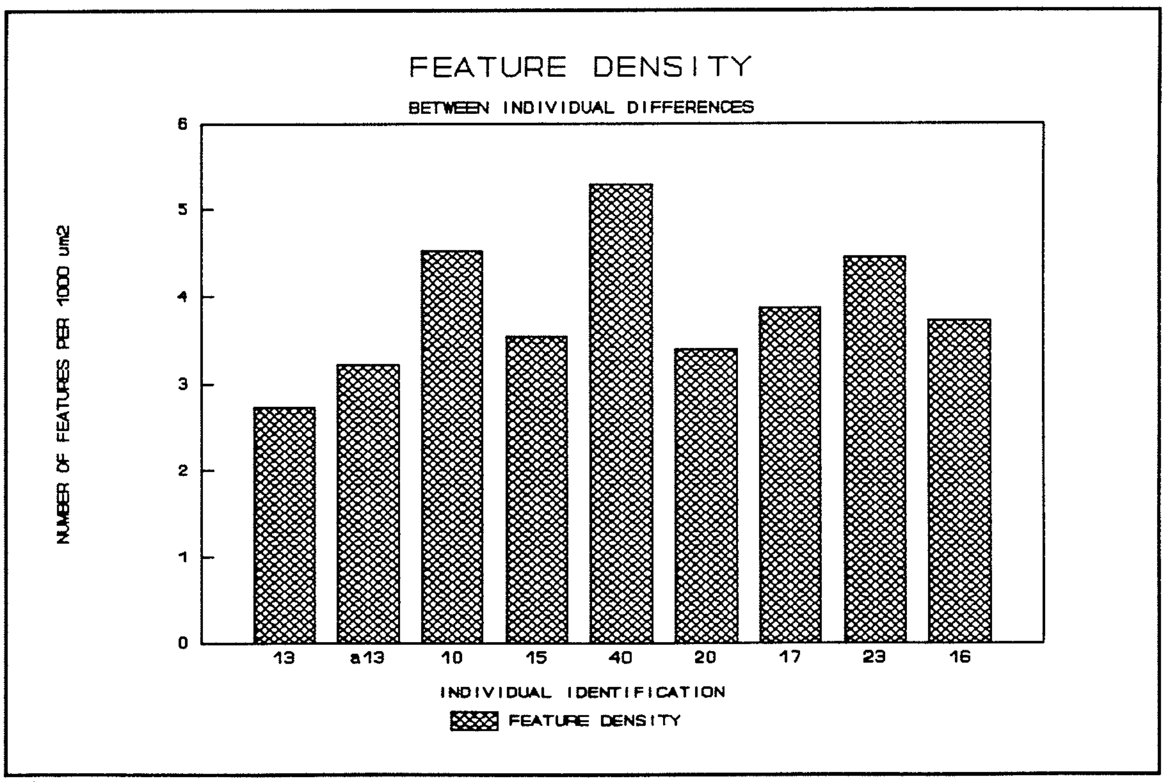


Figure 15d

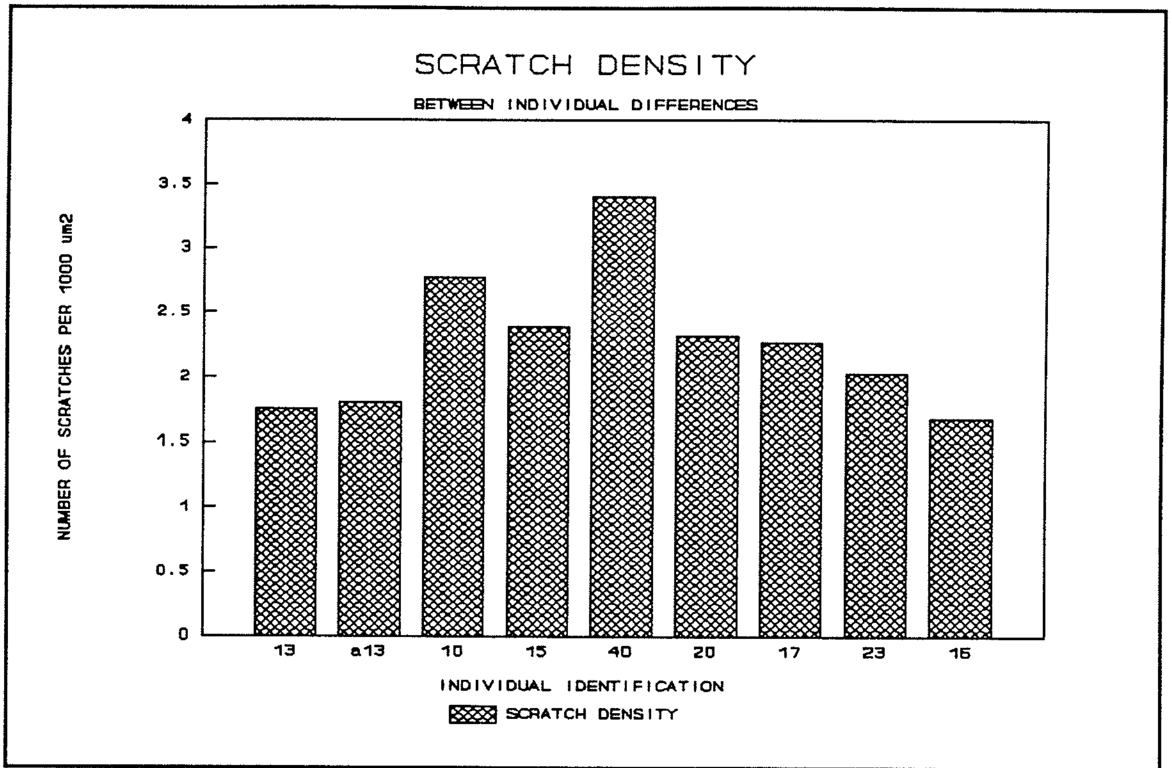


Figure 15e

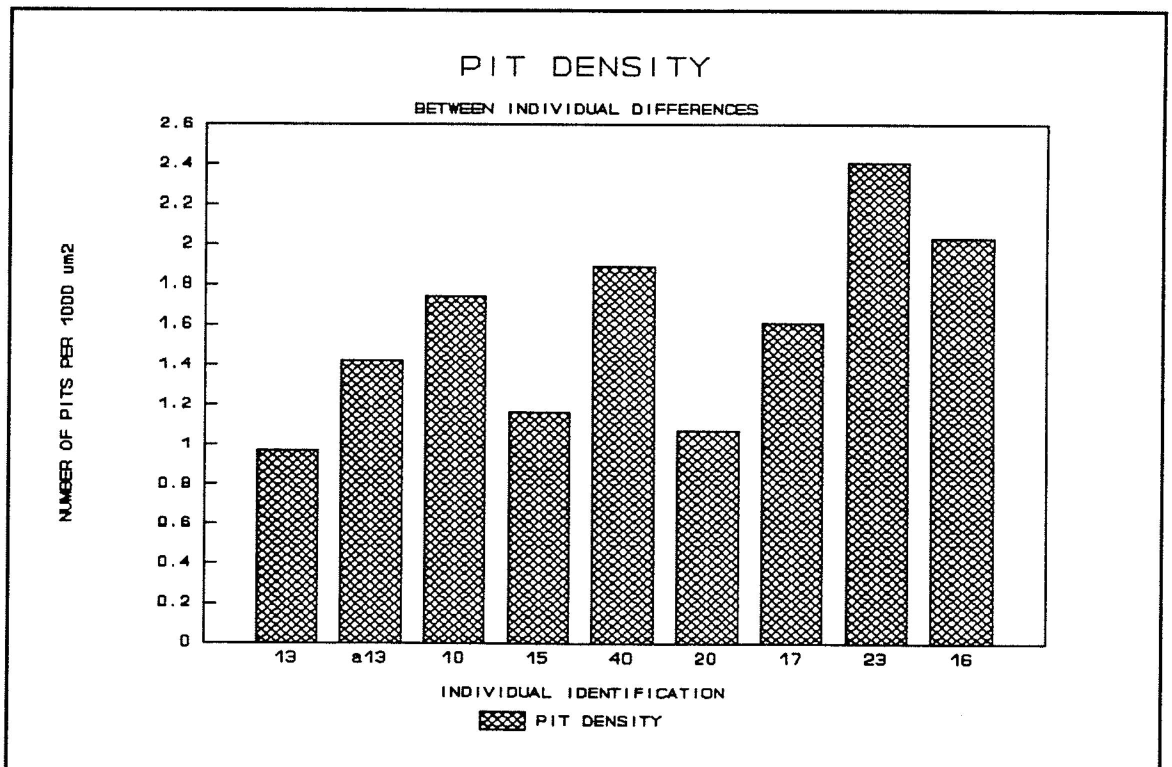


Figure 15f

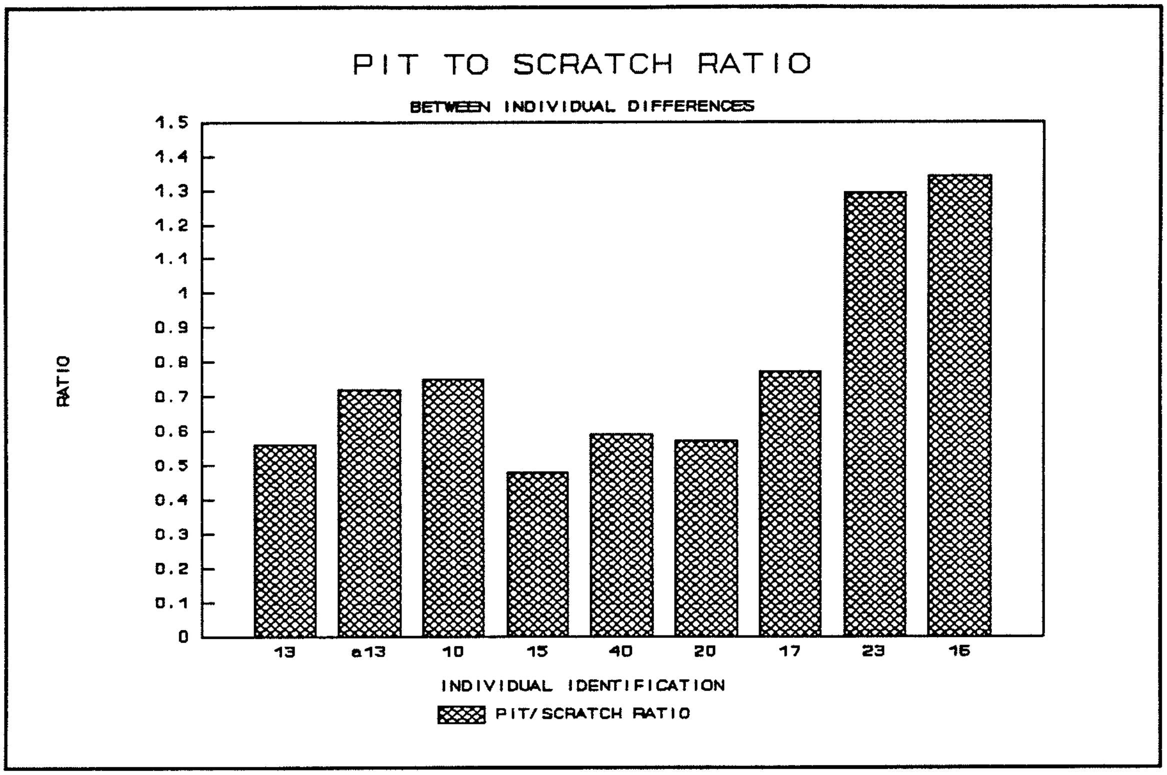


Figure 15g

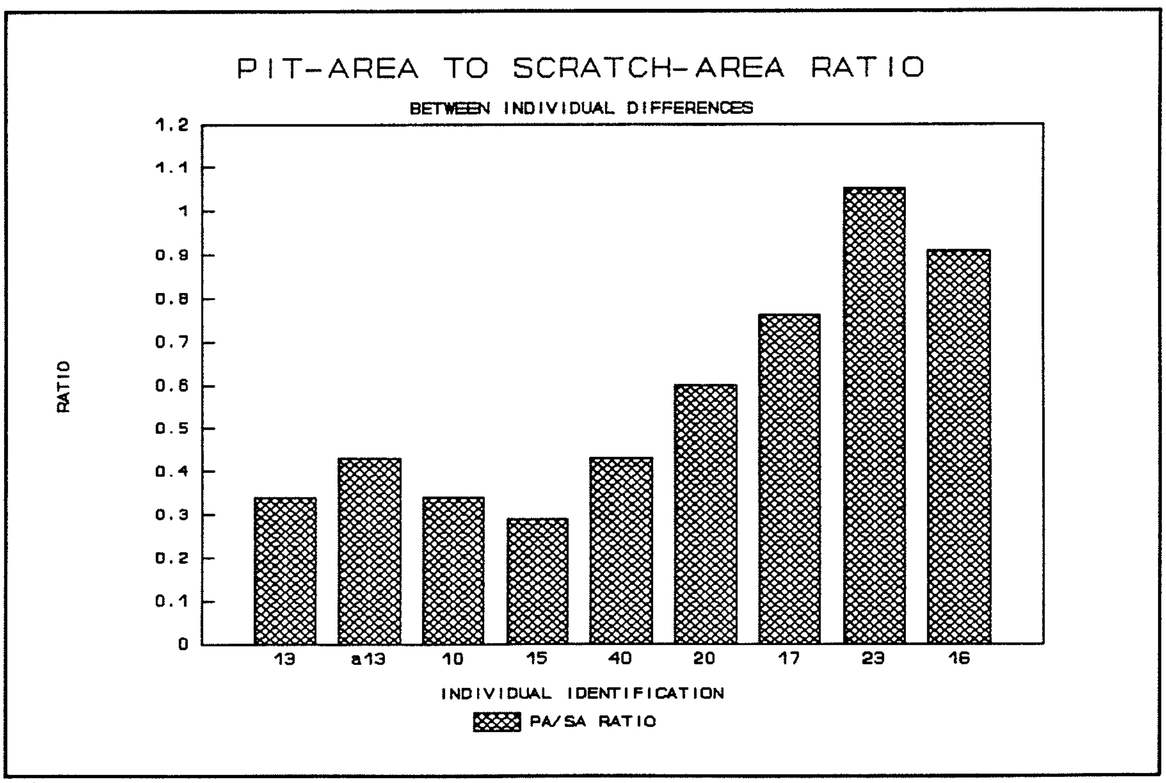


Figure 15h

Figure 16. Separation of Ganj Dareh individuals using two parameters at once.

The use of two parameters at once for individual separations is illustrated by the use of the following graphs. As mentioned in the methods section, the ability to fully assess a microwear pattern depends on not just one parameter. Two or more parameters are required to quantify the size, shape, and number of features that make up a microwear pattern. When two parameters were considered at the same time the separation between individuals become slightly clearer.

Two of these scatter plots, A_Length vs. A_Width, and A_Ratio vs. FD, when considered together, gave us the ability to consider size, shape, and number of features for a comprehensive examination of all characteristics of microwear features. The values and their standard deviations for each of the individuals were plotted.

Figure 16a. Graph of A_Length vs. A_Width of features. The plot of average feature length against average feature width compared both the average size and shape of features between individuals. The greatest differences were found between individuals 13 and the two similar individuals, 16 and 23.

Figure 16b. The plot of average ratio against feature density brings the factor of numbers of features into consideration. This graph also clearly separates out the same individuals as Fig. 16a. In addition, individual 40 appeared to be significantly different than some of the others.

Figure 16c. Traditional pit to scratch ratio and feature density showed a clear separation of individuals 16 and 23 from the rest of the individuals, they appeared to be furthest separated from individual 13 or 15.

Figure 16d. The use of pit-area to scratch-area ratio and feature density shows a similar pattern, but the distinction of 16 and 23 from the rest of the individuals became less clear.

Figure 16. Separation of Ganj Dareh individuals by using two parameters at once.

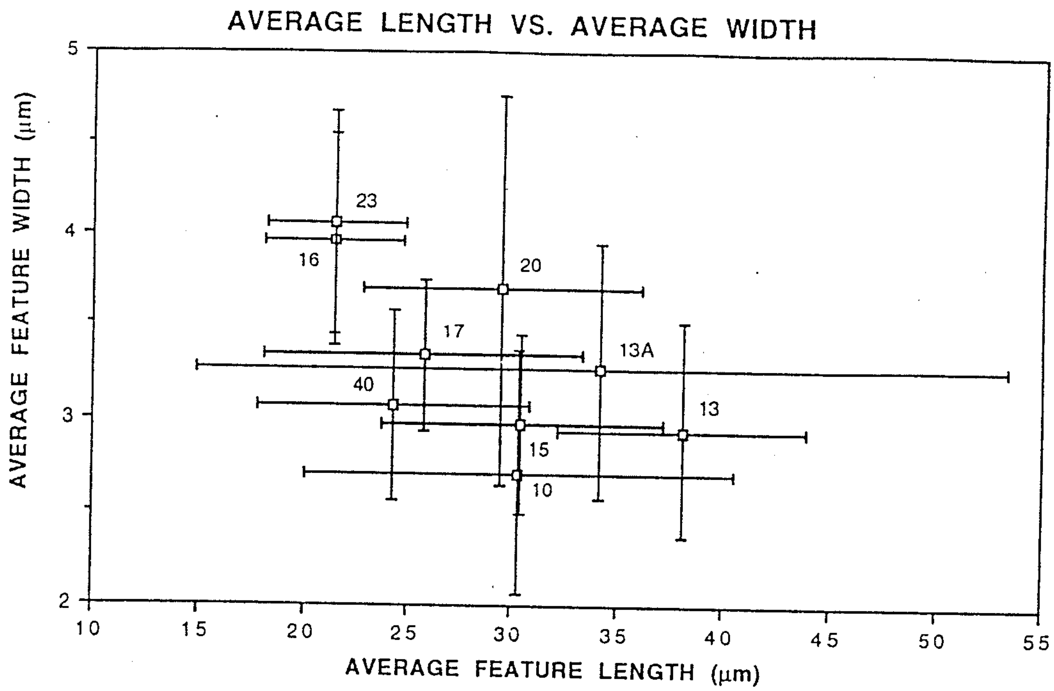


Figure 16a

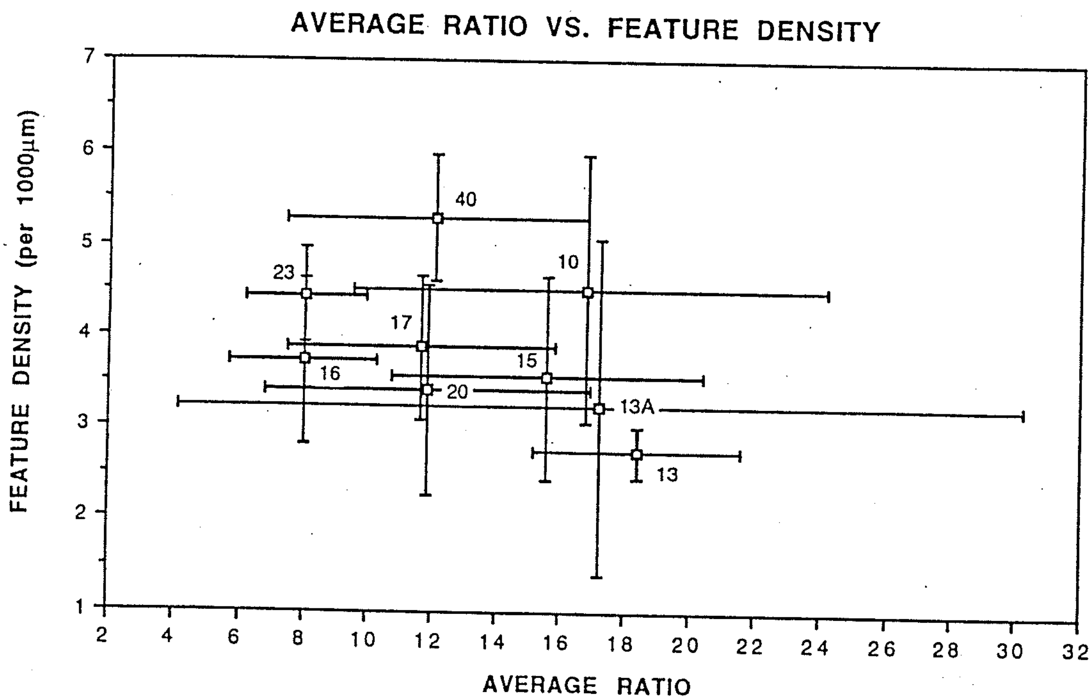


Figure 16b

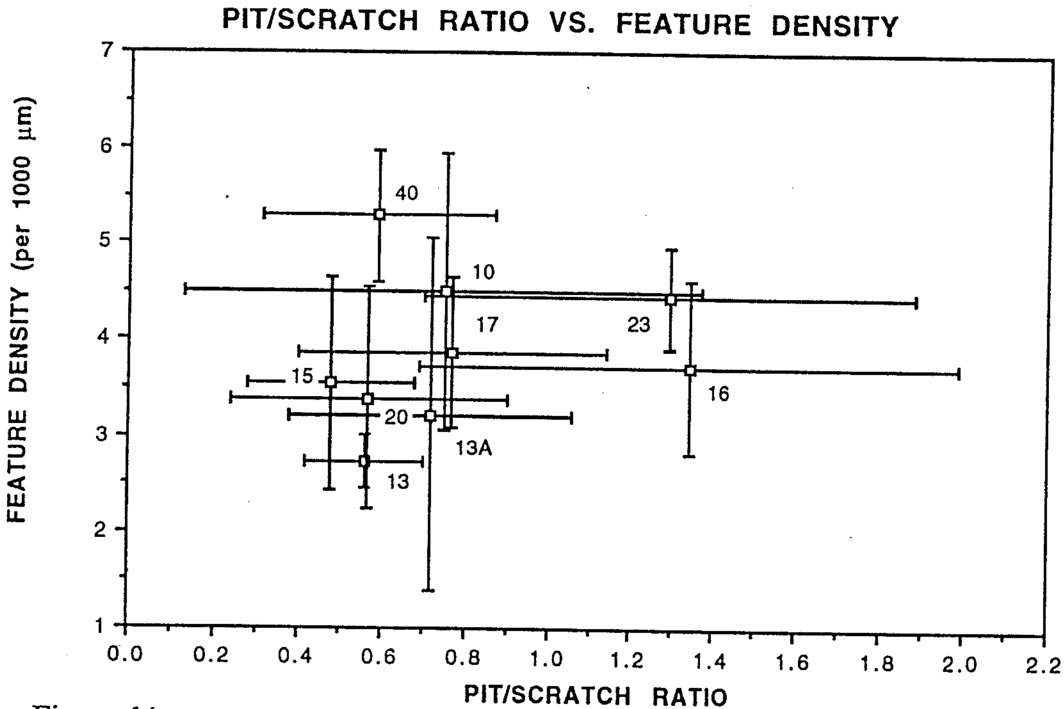


Figure 16c

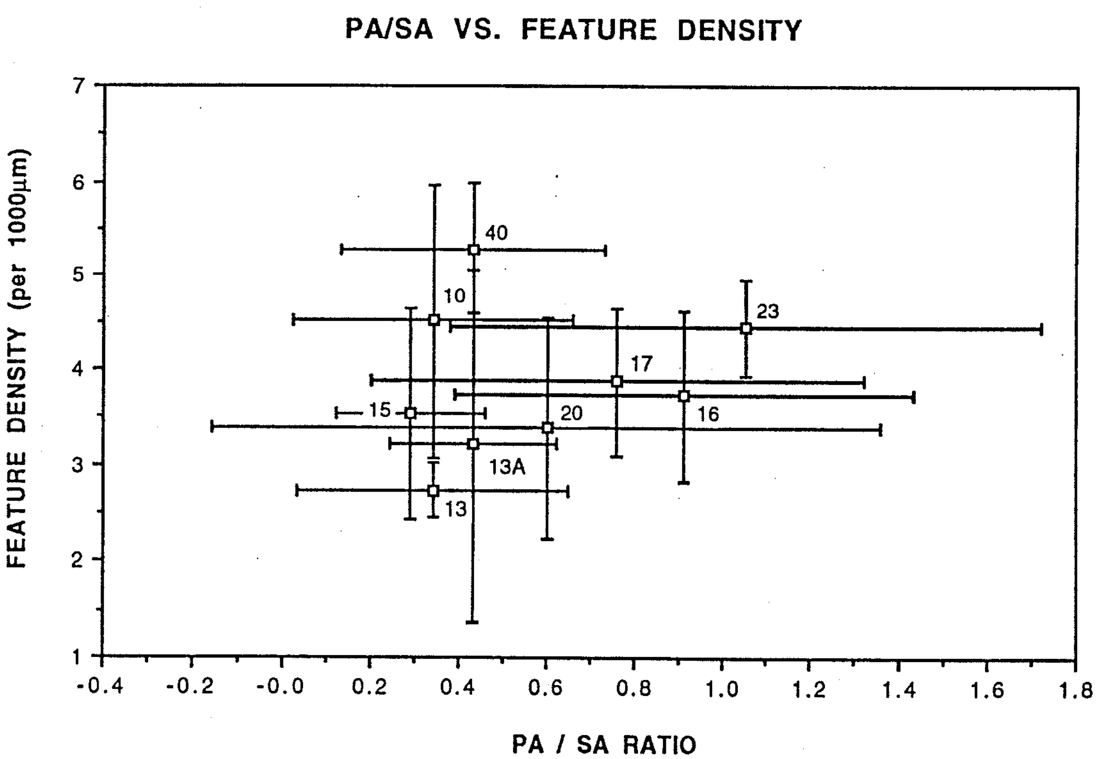


Figure 16d

Figure 17. Representative photomicrographs of microwear patterns found in Ganj Dareh individuals.

SEM photomicrographs taken at 480X magnification of representative individuals from the Ganj Dareh samples are shown here. The individuals selected for this illustration represent the range of different microwear patterns found in the Ganj Dareh samples.

Figure 17a. Photomicrograph of facet 9 from tooth 36 (lower left first molar) of Individual 13. This individual appeared to have a microwear pattern that was predominantly scratches.

Figure 17b. Photomicrograph of facet 9 from tooth 16 (upper right first molar) of individual 16. There appeared to be a much greater percentage of pits present in this photomicrograph than that of individual 13.

Figure 17c. Photomicrograph of facet 9 from tooth 26 (upper left first molar) of individual 23. The microwear pattern found in this individual appeared to be very similar to that of individual 16.

Figure 17d. Photomicrograph of facet 9 from tooth 16 (upper right first molar) of individual 40. The microwear pattern found in this individual appeared to have a pit to scratch ratio that was between individual 13 and 16, but there appeared to be a greater amount of features on this micrograph.

Figure 17. Representative photomicrographs of microwear patterns found in Ganj Dareh individuals.

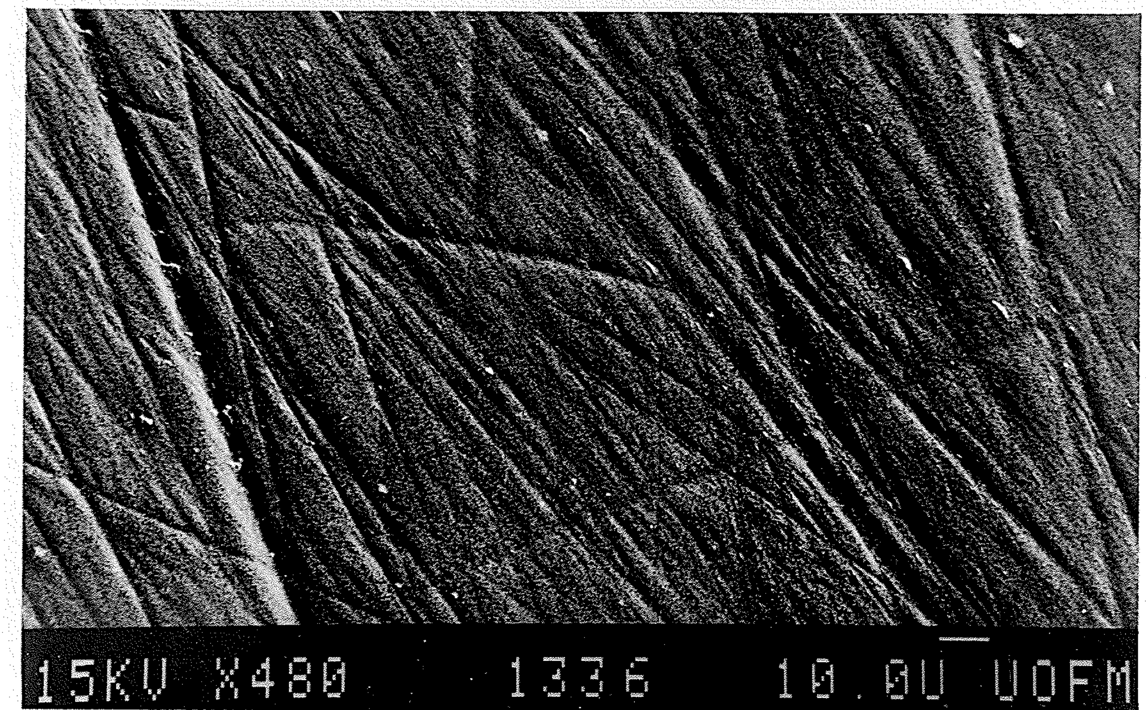


Figure 17a

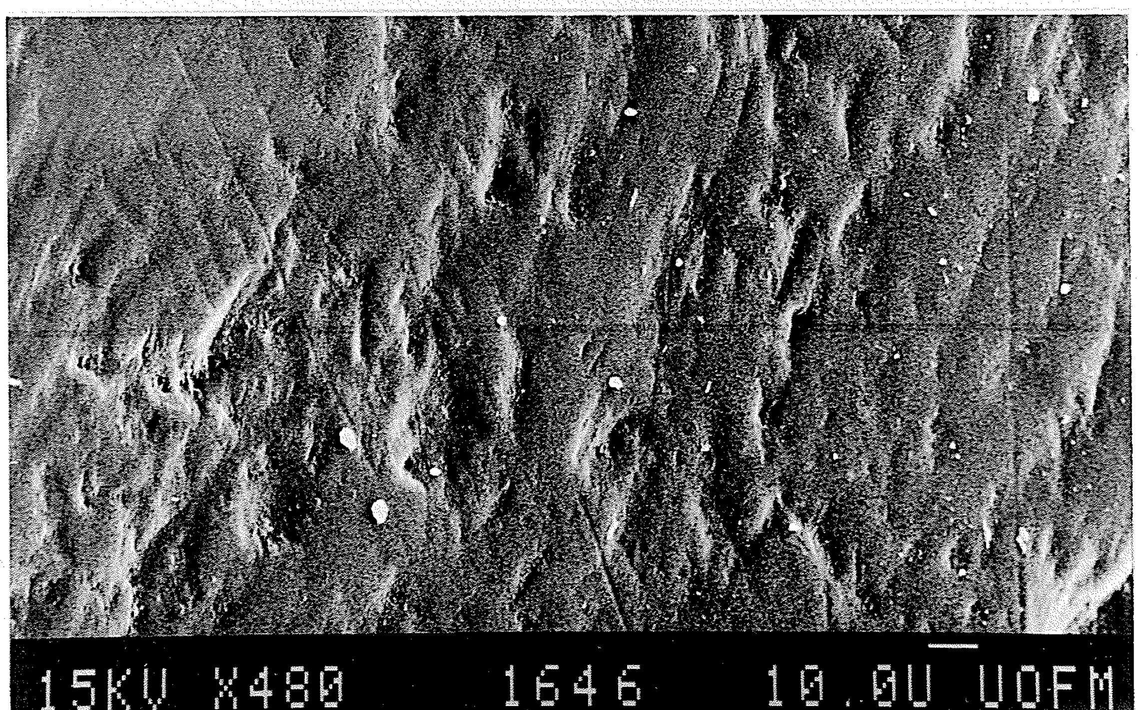


Figure 17b

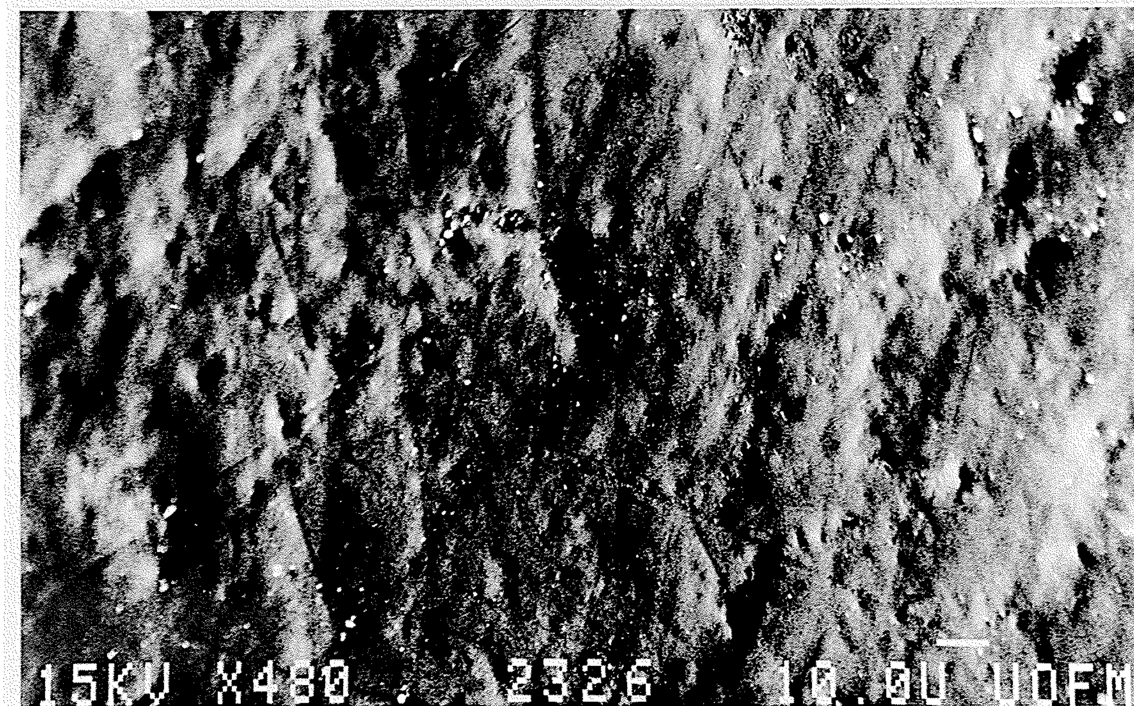


Figure 17c

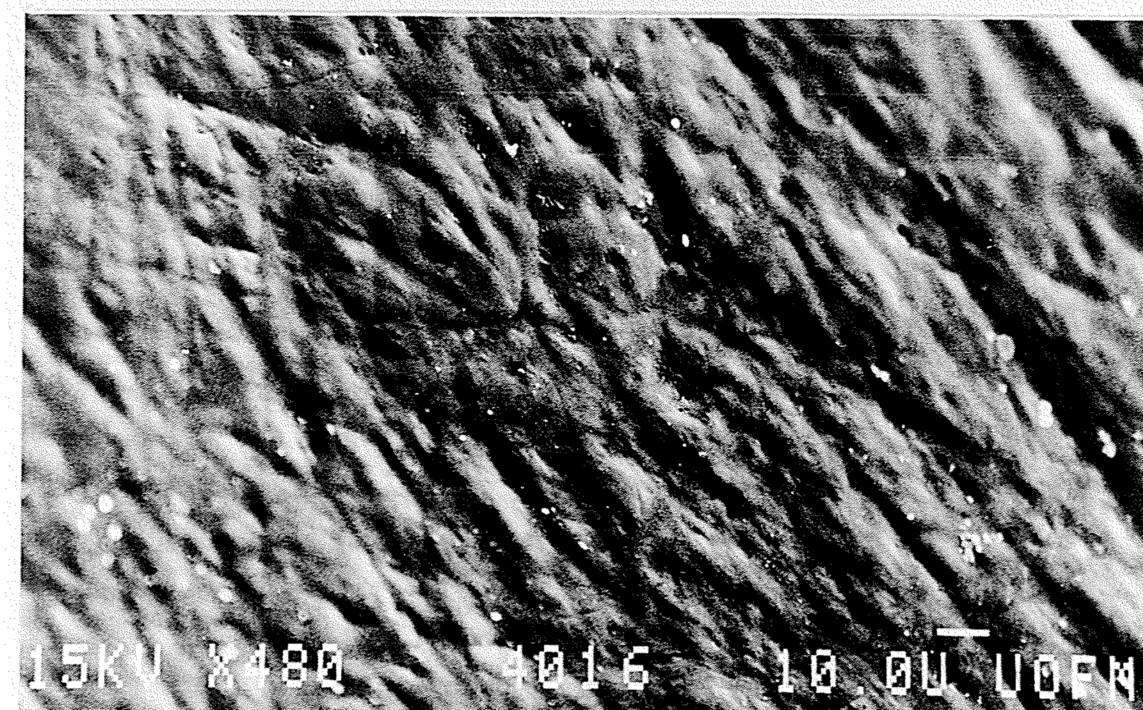


Figure 17d

Figure 18. Dental microwear pattern of Ganj Dareh individuals compared with seven species of primates with known diets.

The microwear pattern of the Ganj Dareh individuals were compared with seven extant species of primates. Data regarding the feature length, feature width, average ratio, and proportional number of pits are presented in graphic form. Data for the primate microwear pattern was obtained from a study conducted by Teaford and Walker (1984). *C. albigena, C. apella, and P pygmaeus* were identified as hard fruit feeders. *C. guereza, A. palliata, and G. gorilla* were identified as soft leaf eaters. *P. troglodytes* was a mixed feeder.

Figure 18a. Graph of feature length vs. feature width showed that Ganj Dareh individuals were closely matched with *P. troglodytes*, a known mixed feeder of hard and soft diets.

Figure 18b. Bar graph of values of average ratio of the Ganj Dareh individuals and the seven species of primates. Again the Ganj Dareh individuals closely matched *P. troglodytes*.

Figure 18c. Bar graph of values of pit density of the Ganj Dareh individuals and the seven species of primates. Pits were defined as any features that had a length to width ratio of <10. The use of length to width ratio of 10 to assess the microwear pattern of species changed the ranking of Ganj Dareh individuals, to a position that was most similar to *P. pygmaeus and C. apella*, both hard object feeders. This was not the only inconsistent finding. The authors reported that the ranking of *A. palliata* also changed when different length to width ratios were used to define pits and scratches.

Figure 18. Dental microwear pattern of Ganj Dareh individuals compared with seven species of primates with known diets.

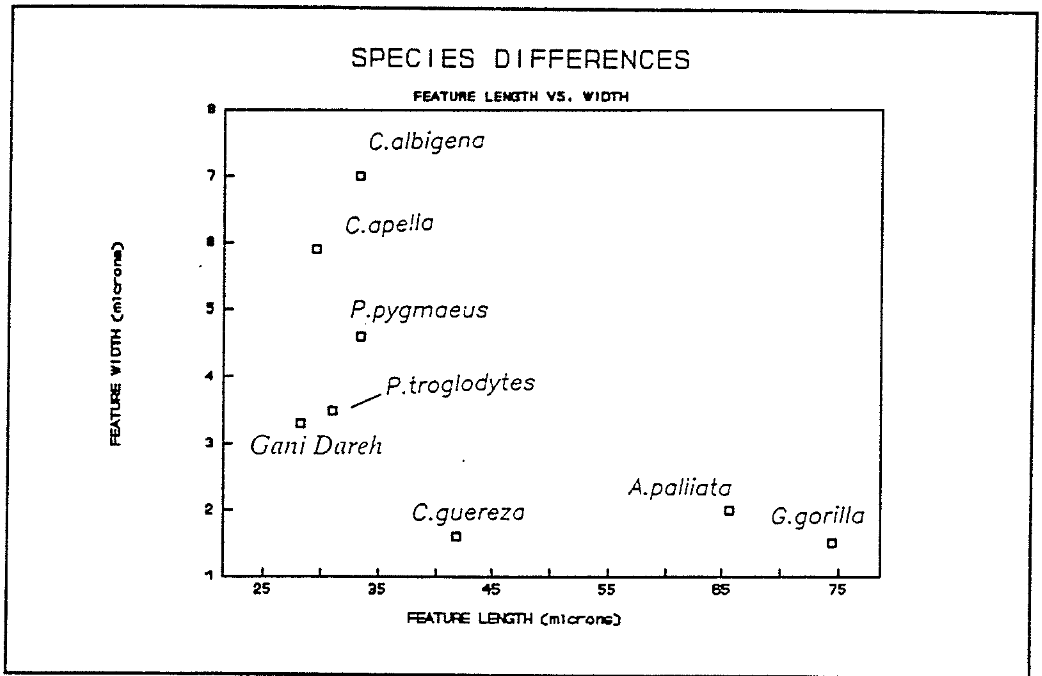


Figure. 18A

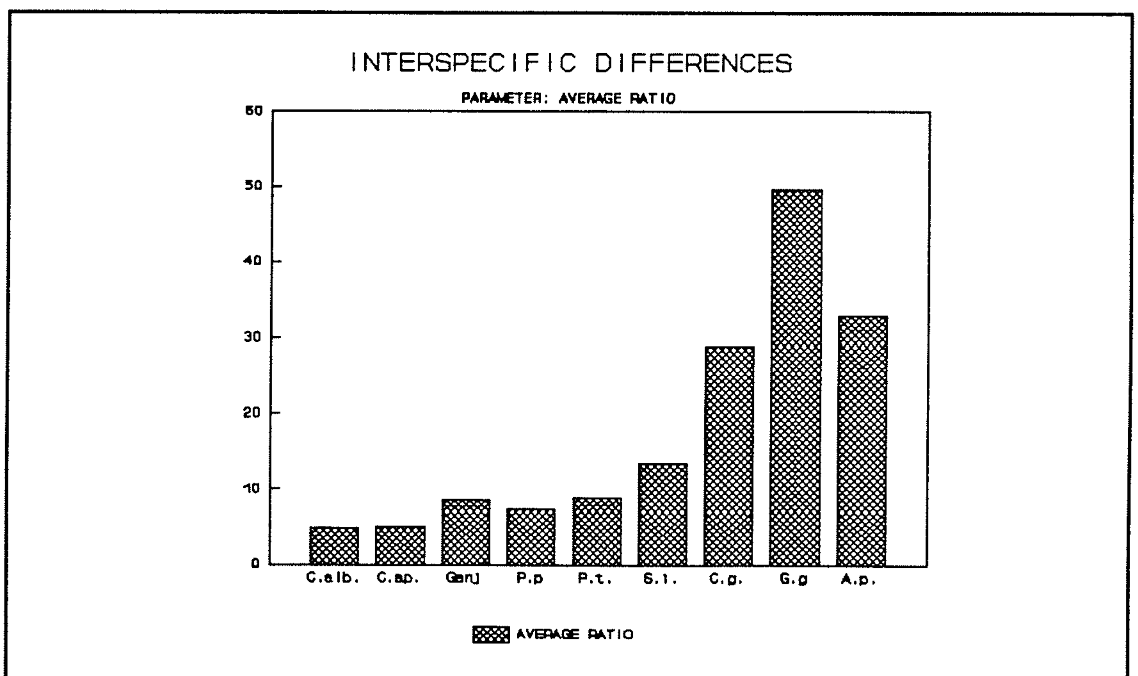


Figure 18B.

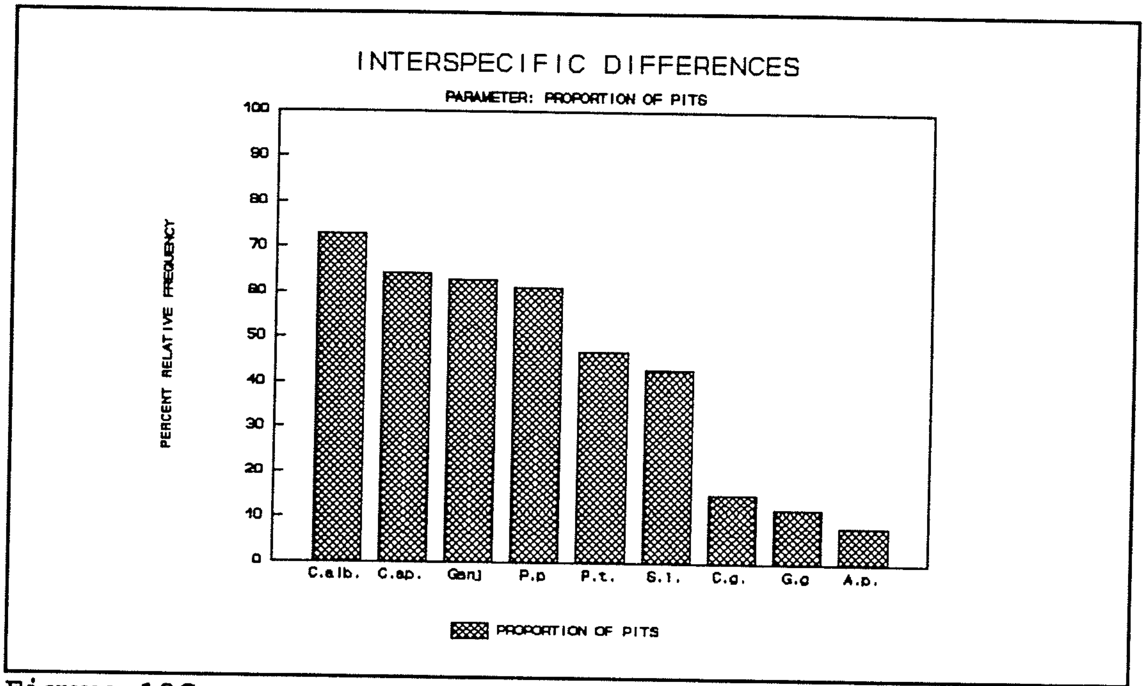

INTERSPECIFIC DIFFERENCES
PARAMETER: PROPORTION OF PITS
PERCENT RELATIVE FREQUENCY
100
90
80
70
60
50
40
30
20
10
0
C.alb.
C.ap.
Ganj
P.p
P.t.
S.l.
C.g.
G.g
A.p.
PROPORTION OF PITS

Figure 18C

Figure 19. Comparison of microwear pattern of Ganj Dareh individuals to other prehistoric human individuals.

The microwear pattern of Ganj Dareh individuals were compared to Mesolithic (★) and Neolithic individuals from Abu Hureyra (□ and ■), as well as modern individuals from Spitalfield and Abu Hureyra (●). The data was obtained from a study conducted by Molleson (*et al.* 1993). The parameters area devoted to pits and total feature area were examined for each of the individuals in this graph. Notice the clustering Ganj Dareh individuals with the modern individuals from Spitalfield and Abu Hureyra.

Figure 19. Comparison of microwear patterns of Ganj Dareh individuals to other prehistoric human individuals.

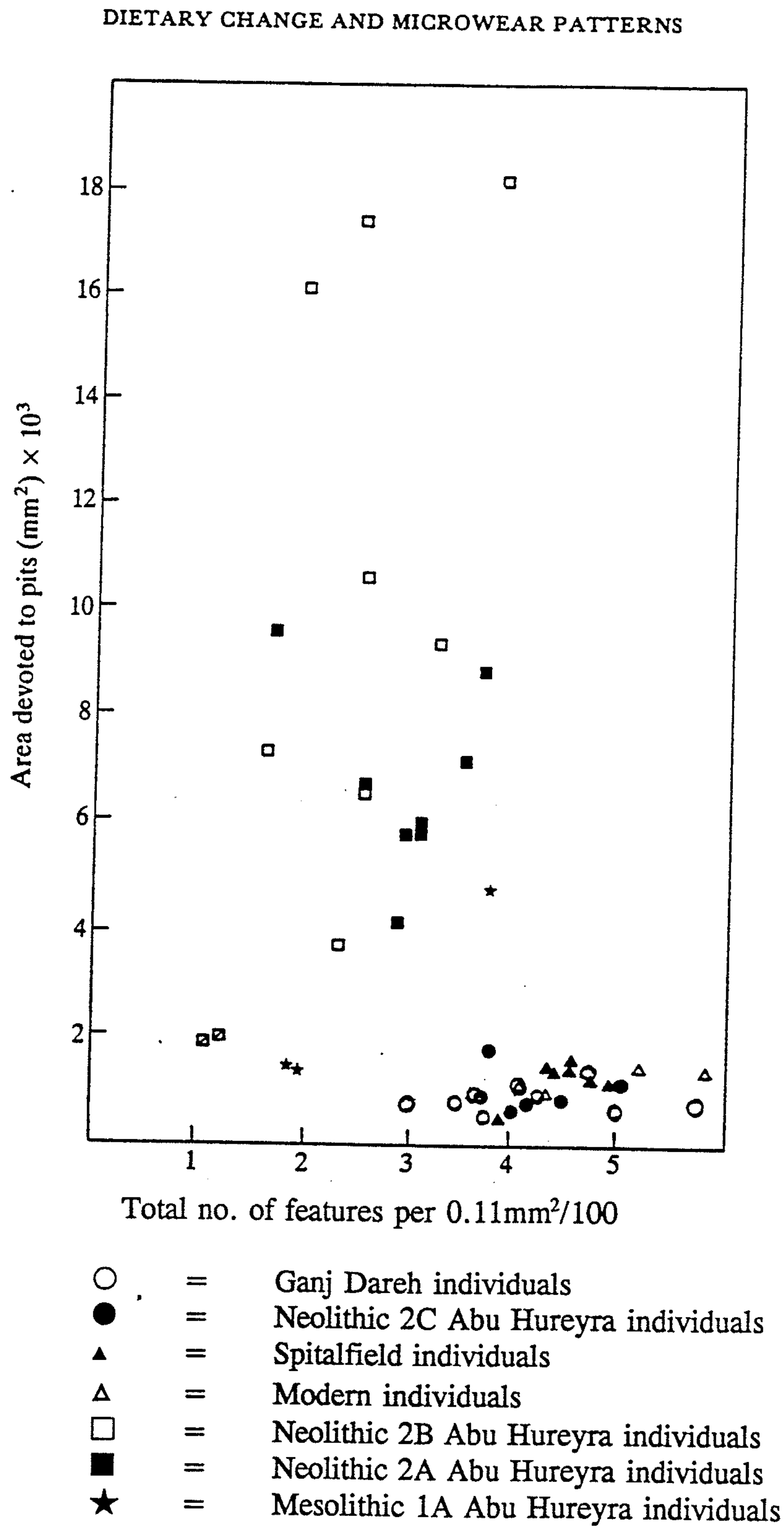


Figure 20. Representative photomicrographs of modern subjects

SEM photomicrographs taken at 480X magnification of occlusal facets from first molars of two modern young adults are shown. Subject A was a 23 year old male, and subject B was a 21 year old female. Impressions of the two first molars on their dominant side (where they had identified as the side they most often chew with) were taken on two separate occasions, eight days apart. Casts were made of these teeth, and representative SEM photomicrographs were taken of the occlusal facets. Evaluations of their microwear patterns were done on a qualitative basis.

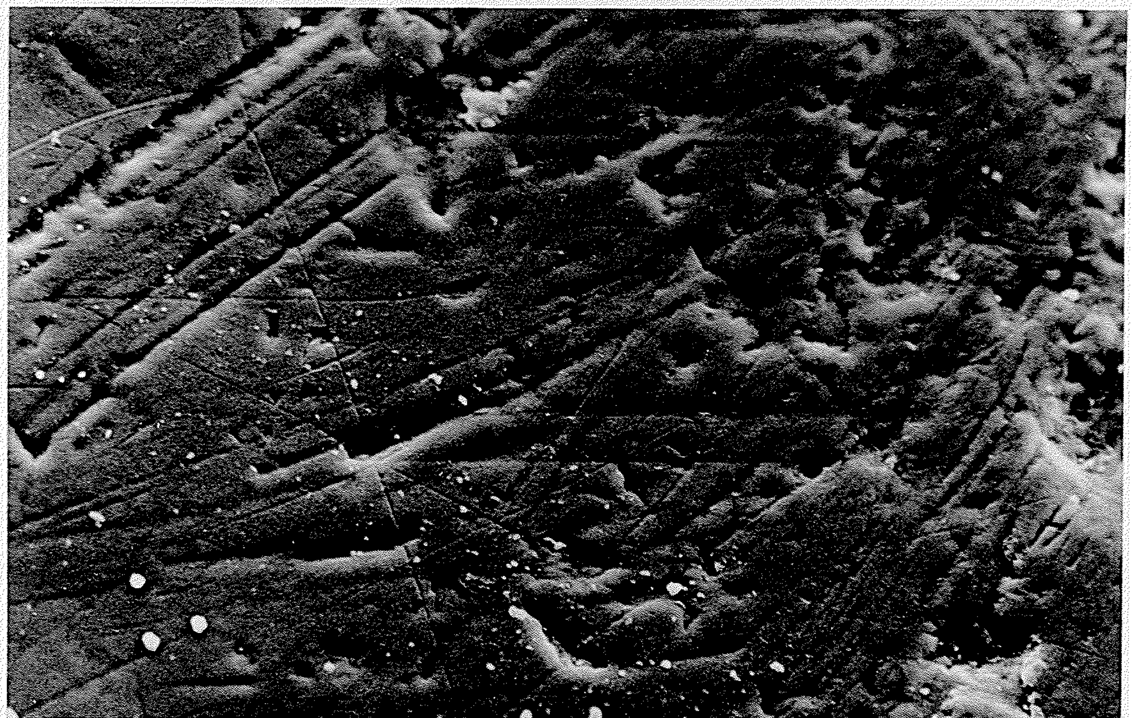
Figure 20a and b. Photomicrographs of cusp tip facets from tooth 26 and 36 (upper left first molar and lower left first molar) of subject A. The microwear pattern appeared to have large deep pits, with parallel sets of very straight scratches that were asymmetric in shape; giving the impression that each set was formed from a single abrasive event. The sets of parallel scratches do not always run in the same direction, and may not be functionally related to the rest of the microwear features.

Figure 20c and d. Photomicrographs of cusp tip facets from tooth 16 and 46 (upper right first molar and lower right first molar) of subject B. The microwear pattern also show large pits on these cusp tip facets.

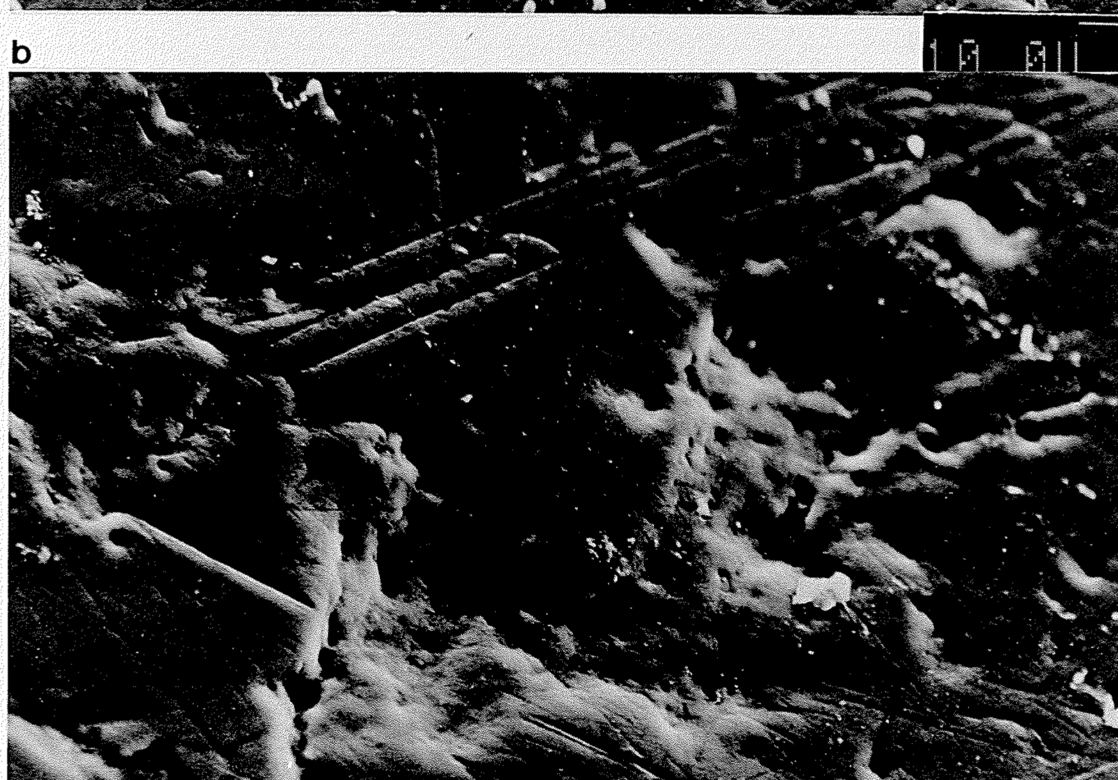
Figure 20e and f. Photomicrographs of facet 9 from tooth 16 and 46 (upper right first molar and lower right first molar) of subject B. The microwear features found in the fossa regions were smaller and contained less pits. Enamel prism relief can be clearly seen on two out of the four photomicrographs taken in this region.

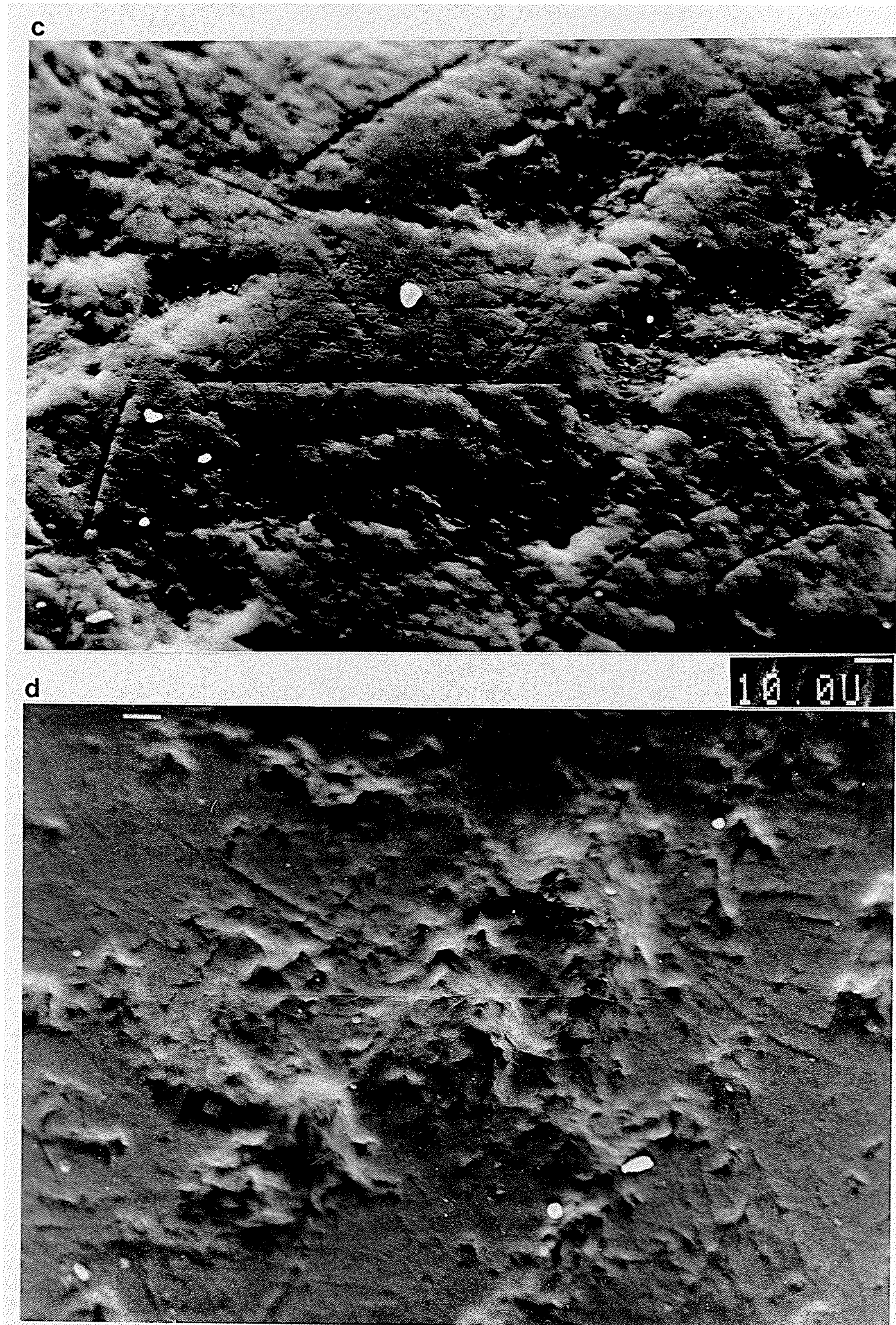
Figure 20. Representative photomicrographs of modern subjects

a

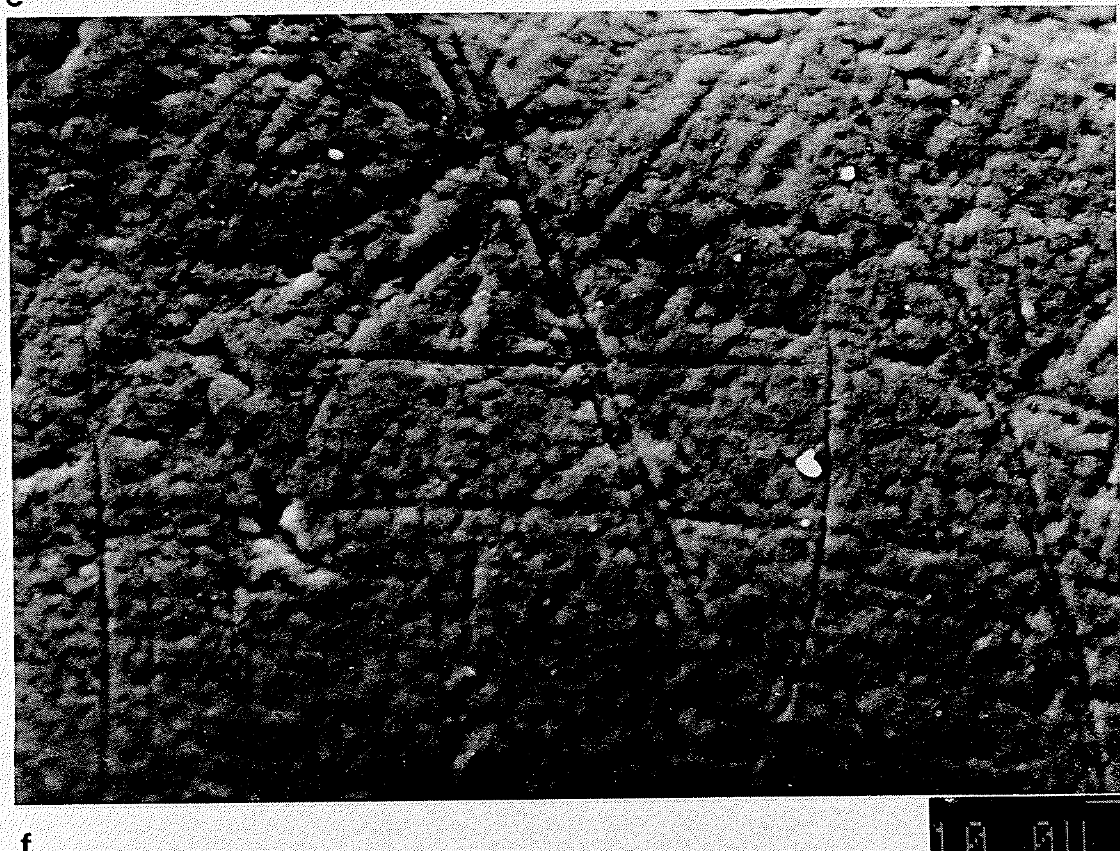


b




c
d
10.0U

e



f

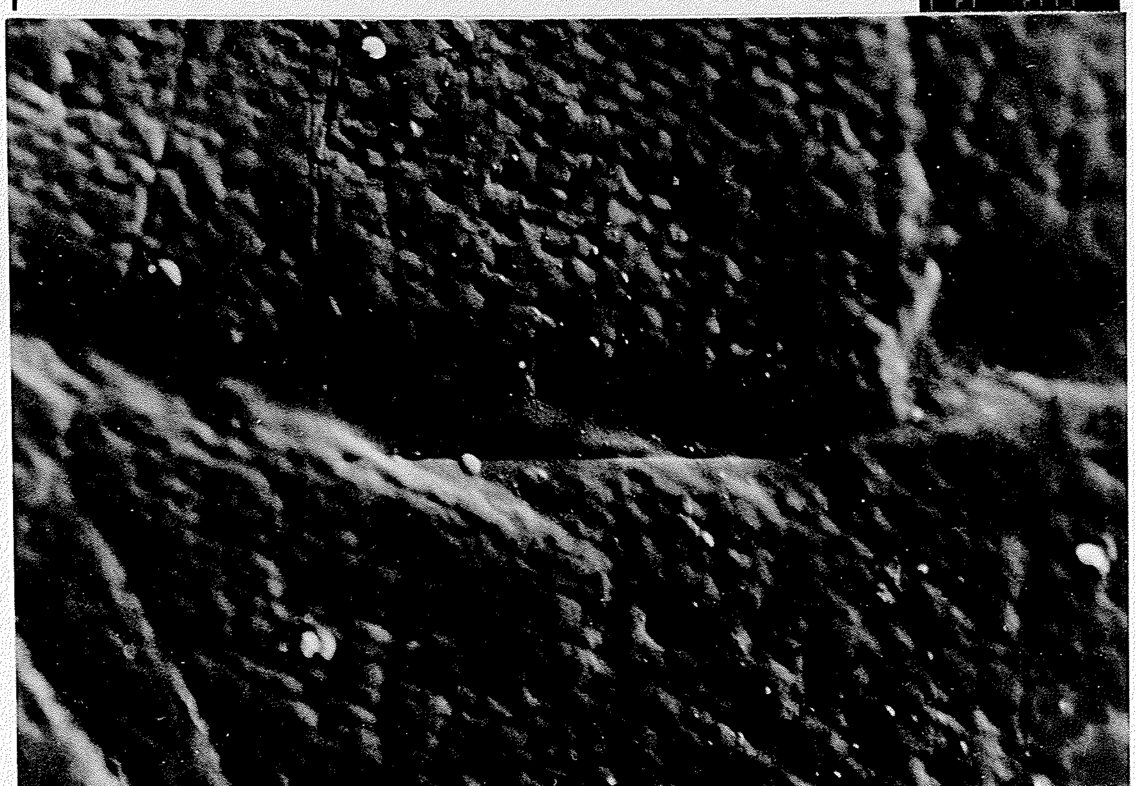


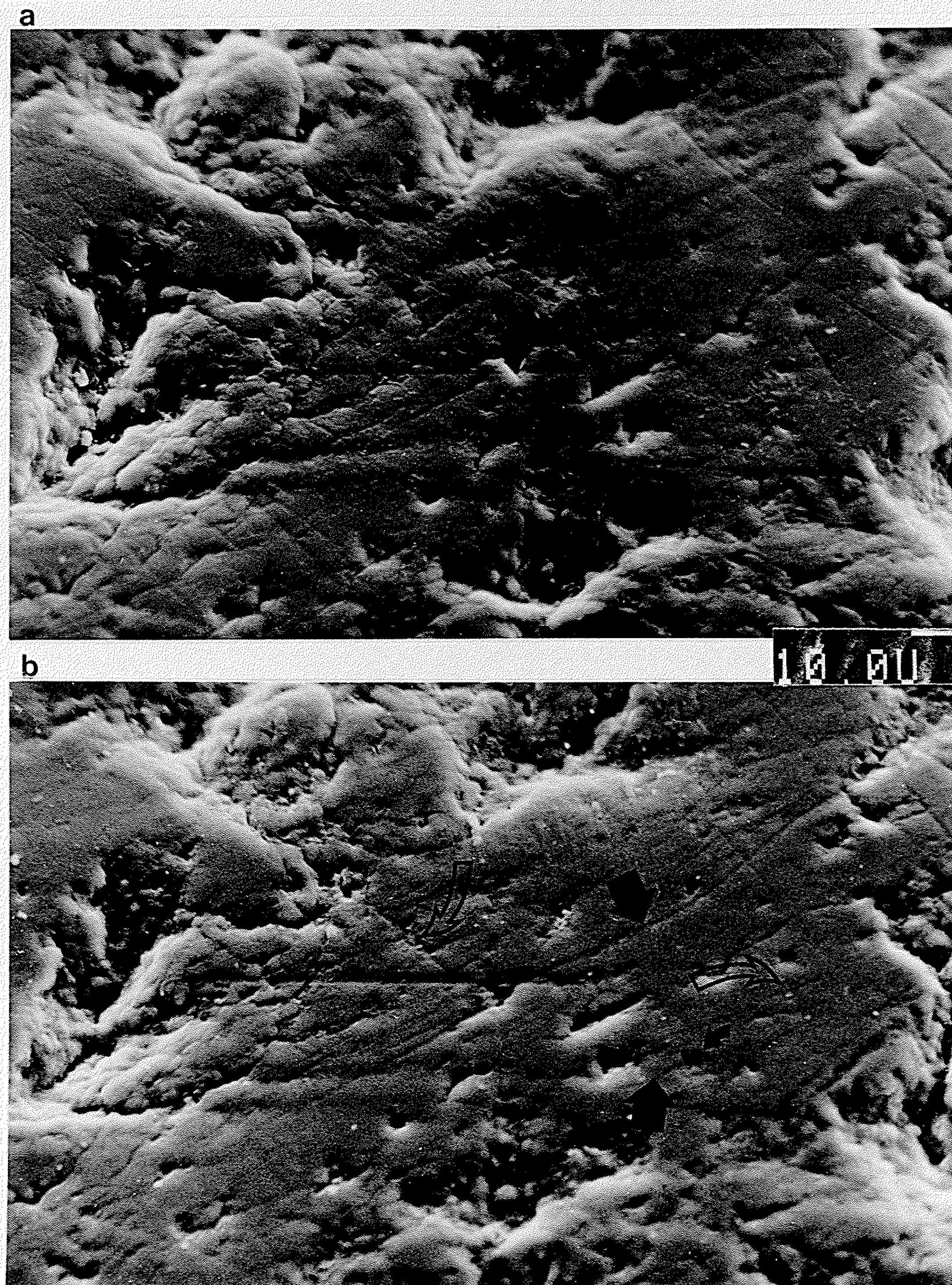
Figure 21. Successive photomicrographs of modern subjects taken eight days apart.

SEM photomicrographs taken at 480X of cusp tip and number 9 facets from the modern subjects. Microwear features on the first of the two successive photomicrographs were identified and counted. New features on the final photomicrograph which were not found on the first photomicrograph were identified and counted. The number of new features formed over the eight days would give an estimated rate of turn over of microwear features found in that facet. Photomicrographs in 21a and 21b are the initial and final photomicrographs from subject A, photomicrographs 21c and 21d are initial and final photomicrographs from subject B. Note the new features found on the final photomicrographs b and d, indicated by the arrows.

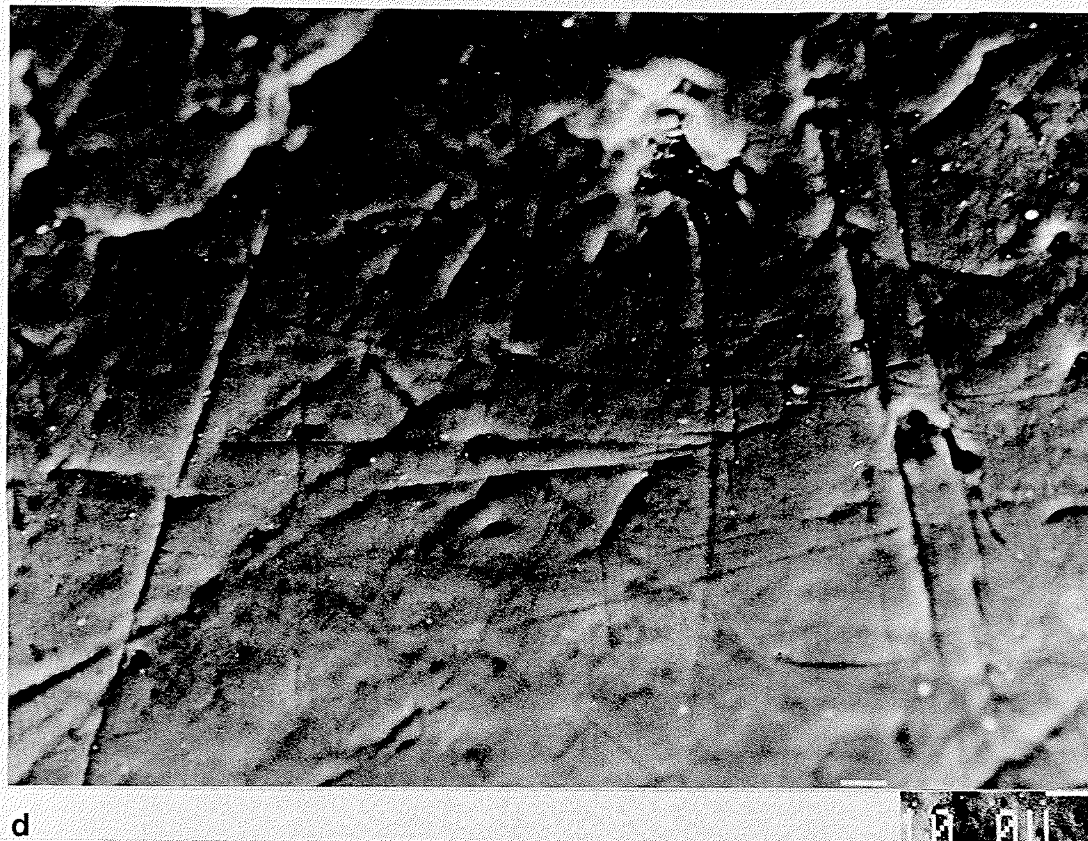
Fig. 21a and 21b. For individual A, the photomicrograph of cusp tip facets were compared. On the initial photomicrograph (Fig. 21a) 149 features were found, with 5 new features found on the final photomicrograph (Fig. 21b). The proportion of new features over 8 days was 0.0336. At this rate, it would take 238 days to replace all the existing features.

Fig. 21c and 21d. For individual B, the photomicrographs of facet 9 region were compared. On the initial photomicrograph (Fig. 21c), 177 features were found, with 2 new features found on the final photomicrograph (Fig. 21d) of the same region. The proportion of new features over 8 days was .0113. At this rate of wear, it would take 707 days to replace all the existing features.

Figure 21. Successive photomicrographs of modern subjects taken eight days apart.



c



d

Table 2. Correlation of Parameters

A new method of microwear analysis involving the area of features was being tested in this study to determine if it would predict the abrasiveness of diet in different individuals. Pearson correlation coefficient analysis was conducted to determine the correlation between the parameters involving feature area and parameters using number of features.

Traditional Parameters:

1) Feature density (FD): number of features per 1,000 μm^2.
2) Pit density (PD): number of pits per 1,000 μm^2.
3) Scratch density (SD): number of scratches per 1,000 μm^2.
4) Pit to Scratch ratio (P/S): number of pits divided by number of scratches in a given field.

New Parameters:

5) Average Length of Feature (A_Length): the average length of the features.
6) Average width of feature (A_Width): the average width of the features.
7) Average Ratio (A_Ratio): average feature length divided by average feature width. This gives the average length to width ratio that will describe the shape of an average feature found in the field, with the assumption that pits and scratches are part of a single microwear continuum.
8) Proportional Area Devoted to Features (FA): sum of all feature areas divided by the field area.
9) Proportional Area Devoted to Pits (PA): sum of all pit areas divided by the field area.
10) Proportional Area Devoted to Scratches (SA): sum of all scratch areas divided by the field area.
11) Pit-Area to Scratch-Area Ratio (PA/SA): pit-area divided by scratch- area in a given field. It is the area equivalent of pit to scratch ratio.

Parameters described above were tested in pairs to determine their correlation. Pairs of parameters that were expected to produce meaningful correlation have been included in this table. The correlation values with their significance level have been presented.

Table 2. Correlation of Parameters

p value ( )

Parameters Tested	P/S vs. PA/SA	PD vs. FD	SD vs. FD	FA vs. FD	PA vs. FD	SA vs. FD	P/S vs. FD	PA/SA vs. FD	PA/SA vs. FA
16	.50 (.02)	.75 (.0001)	.67 (.0005)	*	*	.61 (.002)	*	*	.51 (.013)
26	.78 (.001)	.63 (.0014)	.72 (.0001)	*	*	*	*	*	*
36	.43 (.07)	.78 (.0001)	.91 .0001)	*	*	*	*	*	.65 (.003)
46	.84 (.0001)	.51 (.011)	.65 (.0008)	*	*	*	*	*	*
Overall	.67 (.0001)	.65 (.0001)	.73 (.0001)	*	*	*	*	*	*

Parameters Tested	P/S vs. FA	PA vs. FA	PS vs. FA	PD vs. FA	SD vs. FA	PD vs. SD	PA vs. SA	SA vs. SD
16	*	.80 (.0001)	.57 (.0005)	*	*	*	*	.71 (.0001)
26	*	*	.79 (.0001)	*	*	*	*	.81 (.0001)
36	*	.91 (.0001)	.67 (.002)	*	*	*	.54 (.006)	.49 (.04)
46	*	.54 (.006)	.52 (.009)	*	*	*	*	.77 (.0001)
Overall	*	.68 (.0001)	.62 (.0001)	*	*	*	*	.73 (.0001)

Parameters Tested	PA vs. PD	P/S vs. A_Ratio	PA/SA vs. A_Ratio	PD vs. A_Ratio	PA vs. A_Ratio	A_Length vs. A_Ratio	A_Width vs. A_Ratio	A_Length vs. A_Width
16	*	-.72 (.0001)	-.46 (.03)	-.65 (.001)	*	.88 (.0001)	-.69 (.0004)	*
26	*	-.64 (.0008)	-.51 (.01)	*	.78 (.0001)	.91 (.0001)	-.68 (.0003)	-.45 (.02)
36	*	-.76 (.0003)	-.67 (.0023)	-.80 (.0001)	-.47 (.05)	.91 (.0001)	-.73 (.0007)	-.47 (.047)
46	*	-.76 (.0001)	-.70 (.0002)	.78 (.0001)	.69 (.0002)	.95 (.0001)	-.73 (.0001)	-.64 (.0008)
Overall	*	-.68 (.0001)	-.56 (.0001)	-.69 (.0001)	-.48 (.0001)	.91 (.0001)	-.69 (.0001)	-.48 (.0001)

* non-significant correlation

Table 3. Within individual differences. Two-way analysis of variance

For those individuals with more than one tooth available, within individual differences were tested. By using a two way analysis of variance, involving the interaction of seven different individuals and four different positions of teeth, the top teeth (teeth number 16 and 26) to bottom teeth (teeth number 36 and 46) were compared; teeth on the right side (16 and 46) to their complements on the left side (36 and 46) for within individual differences with respect to all the parameters established below were also compared. In the case of top to bottom comparison, tooth 16 was paired with 46, 26 is paired with 36 in each individual. The side to side comparisons were conducted with tooth 16 paired with 26, and 36 paired with 46 in each individual (Refer to Table 1 for detailed tooth and individuals available for comparisons). The p values for the analysis of variance tests are listed in this table.

Parameters that were tested for within individual differences were:

1) Feature density (FD): number of features per 1,000 μm^2.
2) Pit density (PD): number of pits per 1,000 μm^2.
3) Scratch density (SD): number of scratches per 1,000 μm^2.
4) Pit to Scratch ratio (P/S): number of pits divided by number of scratches in a given field.
5) Average Length of Feature (A_Length): the average length of the features.
6) Average width of feature (A_Width): the average width of the features.
7) Average Ratio (A_Ratio): average feature length divided by average feature width. This gives the average length to width ratio that will describe the shape of an average feature found in the field, with the assumption that pits and scratches are part of a single microwear continuum.
8) Proportional Area Devoted to Features (FA): sum of all feature areas divided by the field area.
9) Proportional Area Devoted to Pits (PA): sum of all pit areas divided by the field area.
10) Proportional Area Devoted to Scratches (SA): sum of all scratch areas divided by the field area.
11) Pit-Area to Scratch-Area Ratio (PA/SA): pit-area divided by scratch- area in a given field. It is the area equivalent of pit to scratch ratio.

Table 3. Within individual differences. Two-way analysis of variance

* p ≤ 0.01

	A_ Ratio	A_ Length	A_ Width	FD	SD	PD	P/S	FA	SA	PA	PA/SA
16 vs. 26	0.68	0.43	0.24	0.96	0.39	0.63	0.28	0.59	0.38	0.38	0.10
36 vs. 46	0.046	0.036	0.04	0.07	0.35	0.12	0.48	0.39	0.69	0.14	0.10
16 vs. 46	0.10	0.08	0.0006*	0.026	0.001*	0.03	0.10	0.64	0.115	0.20	0.06
26 vs. 36	0.45	0.95	0.76	0.65	0.89	0.23	0.17	0.87	0.13	0.48	0.25

* significant difference only found between teeth in individual 20.

Table 4. Between Individual Differences Analysis of Variance

One way analysis of variance was used for the test of between individual differences. The data from all the teeth within an individual were pooled together to represent that individual. The p values for the analysis of variance test are listed in this table.

Sex and age differences between individuals were also examined, for those individuals that have their sex and age identified. Two-way analysis of variance was used to detect sex and age differences. For the age analysis, individuals were group according to child (individuals 12 years old or under) and adults (individuals over the age of 12). There were a total of two individuals 12 years old or under, and 7 individuals over 12 years of age. Sex differences were examined with 2 females and 5 males.

Parameters that were tested for between individual differences were:

1) Feature density (FD): number of features per 1,000 μm^2.
2) Pit density (PD): number of pits per 1,000 μm^2.
3) Scratch density (SD): number of scratches per 1,000 μm^2.
4) Pit to Scratch ratio (P/S): number of pits divided by number of scratches in a given field.
5) Average Length of Feature (A_Length): the average length of the features.
6) Average width of feature (A_Width): the average width of the features.
7) Average Ratio (A_Ratio): average feature length divided by average feature width. This gives the average length to width ratio that will describe the shape of an average feature found in the field, with the assumption that pits and scratches are part of a single microwear continuum.
8) Proportional Area Devoted to Features (FA): sum of all feature areas divided by the field area.
9) Proportional Area Devoted to Pits (PA): sum of all pit areas divided by the field area.
10) Proportional Area Devoted to Scratches (SA): sum of all scratch areas divided by the field area.
11) Pit-Area to Scratch-Area Ratio (PA/SA): pit-area divided by scratch- area in a given field. It is the area equivalent of pit to scratch ratio.

Table 4. Between Individual Differences Analysis of Variance

* p ≤ 0.01

	A_ Ratio	A_ Length	A_ Width	FD	SD	PD	P/S	FA	SA	PA	PA/SA
IND.	0.0001 *	0.0005*	0.0001*	0.0001*	0.0001*	0.0001*	0.0001*	0.19	0.019	0.085	0.008*
IND. and SEX	0.8335	0.7569	0.8591	0.9719	0.8271	0.6332	0.5123	0.6214	0.6548	0.8233	0.8710
IND. and AGE	0.7335	0.4809	0.9872	0.6741	0.9248	0.4112	0.2578	0.6659	0.7712	0.8615	0.8170

Table 5. Ganj Dareh data for each of the between individual parameters.

Statistical analysis could not be conducted to determine whether some or all of the individuals were different from one another due to the unbalanced sampling of individuals. Differences between individual microwear pattern were examined by comparing individual values of each of the parameters.

The values of the eleven parameters for each of the nine individuals are listed in this table:

1) Feature density (FD): number of features per 1,000 μm^2.
2) Pit density (PD): number of pits per 1,000 μm^2.
3) Scratch density (SD): number of scratches per 1,000 μm^2.
4) Pit to Scratch ratio (P/S): number of pits divided by number of scratches in a given field.
5) Average Length of Feature (A_Length): the average length of the features.
6) Average width of feature (A_Width): the average width of the features.
7) Average Ratio (A_Ratio): average feature length divided by average feature width. This gives the average length to width ratio that will describe the shape of an average feature found in the field, with the assumption that pits and scratches are part of a single microwear continuum.
8) Proportional Area Devoted to Features (FA): sum of all feature areas divided by the field area.
9) Proportional Area Devoted to Pits (PA): sum of all pit areas divided by the field area.
10) Proportional Area Devoted to Scratches (SA): sum of all scratch areas divided by the field area.
11) Pit-Area to Scratch-Area Ratio (PA/SA): pit-area divided by scratch- area in a given field. It is the area equivalent of pit to scratch ratio.

Table 5. Ganj Dareh data for each of the between individual parameters.

Standard deviation ( )

	A_ Ratio	A_ Length (μm)	A_ Width (μm)	FD (per $1000\mu m$)	SD (per $1000\mu m$)	PD (per $1000\mu m$)	P/S	FA (per $1000\mu m_2$)	SA (per $1000\mu m_2$)	PA (per $1000\mu m_2$)	PA/SA
10	16.86 (7.32)	30.26 (10.22)	2.71 (0.65)	4.51 (1.45)	2.77 (1.15)	1.74 (1.23)	0.75 (0.62)	0.28 (0.12)	0.21 (0.11)	0.06 (0.06)	0.34 (0.32)
13	18.39 (3.19)	38.05 (5.86)	2.94 (0.58)	2.73 (0.28)	1.76 (0.30)	0.97 (0.13)	0.56 (0.14)	0.26 (0.12)	0.19 (0.04)	0.07 (0.06)	0.34 (0.31)
13A	17.21 (13.05)	34.15 (19.31)	3.26 (0.69)	3.22 (1.84)	1.80 (0.71)	1.42 (1.13)	0.72 (0.34)	0.22 (0.06)	0.15 (0.02)	0.07 (0.04)	0.43 (0.19)
15	15.61 (4.83)	30.44 (6.75)	2.97 (0.48)	3.54 (1.11)	2.39 (0.60)	1.16 (0.61)	0.48 (0.20)	0.22 (0.07)	0.17 (0.04)	0.05 (0.04)	0.29 (0.17)
16	7.97 (2.32)	21.39 (3.33)	3.97 (0.58)	3.72 (0.90)	1.68 (0.57)	2.03 (0.70)	1.34 (0.65)	0.23 (0.06)	0.12 (0.03)	0.11 (0.05)	0.91 (0.52)
17	11.66 (4.20)	25.72 (7.67)	3.34 (0.41)	3.87 (0.78)	2.27 (0.60)	1.61 (0.63)	0.77 (0.37)	0.26 (0.09)	0.16 (0.05)	0.10 (0.07)	0.76 (0.56)
20	11.86 (5.11)	29.48 (6.65)	3.70 (1.06)	3.39 (1.15)	2.32 (1.11)	1.07 (0.39)	0.57 (0.33)	0.29 (0.09)	0.20 (0.07)	0.09 (0.10)	0.60 (0.76)
23	8.01 (1.90)	21.46 (3.28)	4.06 (0.61)	4.44 (0.52)	2.03 (0.46)	2.41 (0.62)	1.29 (0.59)	0.31 (0.05)	0.17 (0.05)	0.15 (0.06)	1.05 (0.67)
40	12.10 (4.72)	24.30 (6.55)	3.07 (0.51)	5.29 (0.69)	3.40 (0.61)	1.89 (0.67)	0.59 (0.28)	0.30 (0.08	0.21 (0.06)	0.08 (0.06)	0.43 (0.30)

Table 6. Ganj Dareh individuals that showed significant differences when parameters Average Length, Average Width, and Feature Density were used for separation.

A new method of microwear analysis, including parameters of average feature length, average feature width, and feature density, has the ability to consider all the characteristics of microwear patterns, and has been shown to possess the best discriminatory ability when used against the Ganj Dareh sample.

Individuals that showed significant differences in their microwear pattern by the use of this method are listed in this table.

Table 6. Ganj Dareh individuals that showed significant differences when parameters Average Length, Average Width, and Feature Density were used for separation.

x indicate significant difference at 1 SD xx indicate significant difference at 2 SD

	10	13	13a	15	16	17	20	23	40
10					x			x	
13					x			xx	xx
13a								x	x
15					x			x	
16	x	x		x					
17									x
20									x
23	x	xx	x	x					
40		xx	x			x	x		

6.0 DISCUSSION

6.1 Method of analysis

Traditional methods of microwear analysis in which separating microwear features into pits or scratches, and then comparing the number of pits, scratches, and total features between different groups of individuals has met with a certain amount of success. When the diet of the groups can be determined, then the microwear pattern of pits, scratches, and number of features of that group have been used to represent that diet. Subsequent groups with unknown diet who present the same pattern of microwear features are then inferred to have the same type of diet. Therefore, it is very important to establish base line data of microwear patterns of different diets with which subsequent groups with unknown diets can be compared.

During the early investigation of microwear patterns, it was found that, at times, microwear pattern of drastically different species produced confusing dietary inferences. Researchers began to look for other factors, besides diet, that will alter the microwear pattern. In the case of anterior teeth, parafunctional requirements such as use of incisors as tools, contributes greatly to the microwear pattern of some species. Molar teeth on the other hand, were rarely used for purposes other than mastication. The differences found can then be attributed to species differences. Species differences can involve morphological and biomechanical differences, such as size and shape of teeth and jaw, relationship of bones and muscle attachment, and masticatory habits, combined to produce a difference in the pattern of occlusion in the chewing cycle, amount and direction of forces developed in mastication, and the amount of chewing surfaces involved. Biological differences may dictate the amount of time and effort placed in processing the food. One study in particular, clearly demonstrated the effect of animal's habit on microwear pattern, involving comparison of microwear patterns of laboratory opossums fed diets with additives of differing abrasiveness (Covert and Kay 1981). The researchers found that they were not able to find any differences between some forms of dietary additives. The assumption made was that one may not be able to distinguish some forms of diets, but the fact that opossums do very little chewing of their food before swallowing might have been the major factor in the inability to find significant microwear differences (Teaford 1988). Other studies on biological, morphological and biomechanical difference between and within species have since revealed the possible significance of age, sex, position of tooth, habits, among factors altering the microwear patterns found. Therefore, one must control for these factors as much as possible, to avoid erroneous conclusions.

The division of microwear features into different categories, such as pits, scratches, gouges, and striations, has been mainly due to the historic evolution of microwear analysis. In the early years of microwear studies, patterns were described qualitatively, with descriptive terms such as surface pitting or striations. When a quantitative method of microwear analysis was developed, the same researchers simply gave these descriptive terms a quantitative value. Pits, scratches, and other categories of features became quantifiable when arbitrary size limits were given for each category. Although some researchers contended that pits and scratches were produced by different agents or processes of tooth wear (Ryan 1979a, 1981), later works demonstrated that the number of pits and scratches found in an individual can vary according to how far back in the tooth row the sample is taken (Gordon 1982). The suggestion was that categories of features were not intrinsically different, but are instead manifestations of differing degrees of shear and compression acting on the abrasive agents which produce the microwear. With this view, microwear features such as pits and scratches are simply opposite ends of a range of microwear features. The decision to categorize features then becomes an arbitrary division of the continuum. This arbitrary decision has been made by all researchers in the field to this day, often with differing opinions as to where the division should be placed. The use of different cut off points for categorizing features into pits and scratches has been found to affect the ability to discriminate some microwear pattern differences (Teaford and Walker, 1984). The prevalent view in the research community appears to be that despite the undesirable effects of this categorization, the procedure continues to be useful, simply having proven to be a significant discriminator between different diets (Gordon 1988).

Despite the fact that more information is being gathered about microwear patterns of different species and their diets, the lack of agreement and inherent unreliability of the categorical approach prevents direct comparisons of works from different researchers. Since pits and scratches are determined by their length to width ratio, at the same time, pits and scratches represent opposite ends of a continuum, then the mean and variation of the feature length to width ratio of this microwear feature continuum should adequately represent the features present. The introduction of average feature ratio as a parameter in this study is meant to simplify and make more consistent the method used for microwear pattern comparison.

The count of microwear features has been used partly out of convenience. Due to the large number of features present and the number of fields required to analyze the data of a very small sample, the calculations required to analyze anything but the number of features has been prohibitive. If one examines the microwear pattern and considers which parameters would be important in determining the diet, one quickly comes to the realization that it is not only the number and shape of features that are important. The size of features has a dramatic impact on the qualitative and possibly quantitative analysis of microwear pattern. When microwear patterns are used for dietary inferences, it is the hardness or the abrasiveness of the diet that is reflected in the features. The harder the diet, the more crushing or grinding is required; actions that require greater amount of vertical forces, or compression, on the food particles and the occluding surfaces of teeth. Greater amount of vertical forces acting on the abrasive particle by occluding surfaces would presumably result in shorter and deeper scars on the enamel. Therefore, the amount and size of the abrasive particle and the amount of force required to break them down would determine the size, shape, and number of features. The count of features, and categorization of features into pits and scratches, only measure the amount and, partly, the shape of microwear features. By neglecting the size factor in quantitative microwear analysis, one cannot adequately differentiate microwear patterns that only differ in their size of features. Patterns with the same number and shape of features, but very different in size, would not be easily mistaken to be the same by visual inspection, since the amount of destruction of the enamel surface would be drastically different. The size of features can only be determined if we also consider the proportion of feature area.

In this study, an attempt has been made to develop methods that will reduce the time required for analysis, and correct inadequacies of an analysis based only on the count of features. First, an automated image analysis method has been attempted, whereby all the features from an SEM image can be quantified by an image analysis system, reducing the amount of time required to do microwear analysis. This phase of the study was not completely successful. The features found on the SEM photomicrographs could not be successfully identified by the image analysis system employed for this study. The major stumbling block was the lack of consistent contrast between the features of interest and the background surfaces. Normally one identifies depressions on a two dimensional image by the change in contrast found between the surface and the depression. Unfortunately, as with many biological specimens, there was a great deal of variation in what is being looked for. In this case, the differences between the contrast of features and the normal surface were often overshadowed by the variation in shade of the normal surface alone. Compounding the problem, the overlapping of multiple features made the identification of each feature a real problem.

Although the search for an automated image analysis system was not entirely successful, a semi-automated system was developed. This system, as described in the methods section, required the identification of features by an operator. Due to the high magnification and sharpness of the SEM images captured on the photomicrographs, the large high definition monitor used to display the image, and the ability for the system to artificially enhance the image contrast, accurate identifications of both the length and width of features were possible. The feature data could then be processed by the computer to produce the final data used in microwear comparisons. Although the system still requires a human operator to define the features, the handling and manipulation of data by the computer has greatly increased the speed at which data can be processed.

The second problem this study tried to address was the inadequate consideration of feature size in microwear analyses. In order to accurately quantify the amount of surface area occupied by microwear features, the area of each features has been estimated. The area parameters considered in this study have been chosen for their possible significance in detecting microwear differences. The proportional area is the counterpart to feature density; providing a true measure of how much area is involved with features. The proportional pit area, proportional scratch area, and pit-area to scratch-area ratio do the same for pit and scratch counts. Although one might argue that the identification of pits still relies on the arbitrary categorization of features, their use in this study was intended to provide area parameters in a form that can be directly compared to traditional number parameters, to assess their significance in microwear studies.

Theoretically, in order to take size, shape, and number of features into consideration, more than one parameter would be needed. Researchers have often used pit density or pit to scratch ratio to estimate the shape of features, and feature density to estimate the number of features. This study has used many parameters to determine all three factors. Proportional area of pits, scratches, and total features have the ability to account for both their size and shape. Pit-area to scratch-area ratio do not directly measure size, but it is a ratio that is based on the size of pits and scratches. The average length and average width of features are linear measurements in themselves, but when considered together, they will give the average size and shape of microwear features. In addition, average length and average width, when considered together with feature density, will include all the important factors of size, shape, and amount in a microwear analysis. Average ratio and feature density can replace pit to scratch ratio and feature density, in considering the shape and amount of microwear patterns; the addition of proportional area of features to either of these parameters can give the third dimension of size in considering the total equation.

The purpose of including all these parameters that can potentially give the similar comparison results was to find a combination of parameters, among all the parameters used, that will give the best discrimination of microwear patterns between different individuals. The best combination of parameters will then constitute the development of a new method for microwear analysis.

6.2 Ganj Dareh material

6.2.1. Microwear Analysis on a Continuous Scale: Test for Validity

The present study attempted to establish the continuous nature of microwear features, and present a method that will truly reflect this nature. As the results indicate, the distribution of feature length to width ratio appears to be continuous in nature. There were no plots that demonstrated multiple peaks around different means, the presence of which would suggest two or more types of features. Distribution of feature ratios possessing only one mean and variation thus demonstrated, strongly suggest that they were continuous in nature. The implication is that there is no real need to produce artificial categories of pits and scratches. The feature length to width ratio will adequately describe the average feature shape. Any change in the number of "pits" will be reflected in the change in average ratio. In fact, the average ratio would be more sensitive to a general trend of feature shape changes, since an increase in the number of "pits" will only occur when the feature length to width ratio drops below 4, whereas the average ratio considers all the features at once. Therefore, average length to width ratio has the potential to be the most significant measure in a microwear analysis of diet.

6.2.2. Correlation of Parameters Used in Microwear Analysis

Correlation analysis of all the parameters used in this study was intended to identify the parameters that give similar results when used in microwear analysis. One of the goals of this project was to suggest new parameters, specifically the area of features, that will give better discrimination in diet interpretation. The parameters involving feature area were compared with parameters using number of features to identify their correlation. A high correlation of area parameters to number parameters would suggest little or no difference in the results when either one is used in microwear analysis.

Overall, there appears to be a weak positive correlation between the parameters that measure the number of features with those that measure the area of features. The weak positive correlations are significant in reflecting the inability of feature numbers to truly predict the amount of tooth surface covered by features. At the same time, the new parameters using feature area have the potential to discriminate microwear patterns in a similar way as the traditional parameters.

The correlation between pit to scratch ratio and pit-area to scratch-area ratio reflects the overall result of a weak positive correlation between number parameters and area parameters. Since pit to scratch ratio has traditionally been the most important parameter used in microwear studies, it was encouraging to see this kind of correlation result for the reason mentioned above.

Average length to width ratio was a new parameter introduce in this study. It is a non-categorical parameter that reflects the continuous nature of microwear features. It shows good correlation with pit to scratch ratio and lower correlation with pit-area to scratch-area ratio (both correlations were significant at $p=.0001$). Again, the correlations indicate a potential for this parameter be a significant microwear pattern indicator. Average ratio, by itself, does not indicate how much of the tooth surface is involved in microwear features, it just indicates what type of features are present. Therefore, it was expected to show less correlation with parameters involving feature area.

Gordon (1982) indicated in her study of chimpanzees that scratch density stays relatively uniform and it was the pit density that varied. This was thought to determine the pit to scratch ratios found on different facets. In this study, both pit density and scratch density have a positive correlation with feature density. When considered together with the fact that pit to scratch ratio does not have a correlation with feature density, one can conclude that, in this sample, both pit and scratch density can vary to determine the final pit to scratch ratio. The different conclusions found between Gordon's study and this study may reflect sample differences. In Gordon's study, she chose only adult chimpanzees, for whom she had controlled for age and sex. Gordon did find that a change in feature density was reflected in the age of her sample. In the present study, with a small sample size, there were individuals from the age of six to fifty. The sample size was too small to control for age and sex.

The proportional area of features also had a positive correlation with proportional area of pits and proportional area of scratches. The pattern was similar to those found for parameters involving number of features.

Scratch density showed positive correlation with proportional area of scratches. The high positive correlation indicates that scratch size was reasonably uniform in this sample. One of the reasons for this uniformity may be due to the field size obtained at 500X magnification. As Gordon (1988) has found, microwear analysis conducted at high magnification may exclude larger scratches from being included in analyses. Any features that are longer than the field length will be truncated, at a frequency of up to 10% of the total number of feature, as found by Gordon (1988). The restriction in the size of scratches that may fit inside the chosen field area can prevent larger scratches from being included, artificially creating uniformity in scratch size.

Pit density and proportional pit area show no correlation with each other. This finding contradicted Molleson's finding that fewer pits mean larger pits. Nevertheless, our findings still indicate that the number of pits was not a good indicator of how much of the field area was covered by pits; again illustrating the inadequacy of pit numbers to predict how destructive the diet can be. The uniformity in scratches in different individuals indicate that scratches were not sensitive to the change in size of abrasive particles or the forces acting on them. Pits, on the other hand, varied much more in their size. What might be concluded from these results were that pits were more sensitive than scratches to the abrasiveness or forces acting on the abrasive particles of a diet.

The correlation of parameters does not give any indication as to which parameters were the best indicator of diet. Since parameters such as pit to scratch ratio and feature density have been used effectively in traditional methods of microwear analysis, their correlation with the new parameters served the purpose of allowing the assessment of the potential usefulness of the new parameters chosen. As the correlation results have indicated, there was an overall positive correlation between traditional and new parameters. Some of the reasons for correlation, or lack thereof, have been discussed, allowing for postulation of characteristics of microwear feature formation. The true test of the discriminative power of these parameter in diet analysis may only be demonstrated when the two methods are compared together on a known sample.

6.2.3. Within Individual Differences

Researchers studying microwear patterns between different individuals have often used only one tooth from each individual for their analysis, with the general assumption that microwear pattern does not vary within an individual. Part of the reasons for this assumption was that microwear analysis has been recognized as an extremely time consuming endeavour, compounded by the restricted availability of complete and balanced samples when working with archaeological material. In order to obtain a large enough representation of the population, many of the researchers have opted, or been forced by availability of samples, to utilize only one tooth to represent each individual. The importance of controlling for position of tooth on the tooth row has been demonstrated previously (see literature review), but the effect of which quadrant the tooth comes from has not been explored. The basic assumption of uniformity in molar microwear pattern appears to be sound; after all, all the molars will experience the same abrasion once the food enters the mouth, and biomechanic considerations should be equal if the teeth are from the same position in the tooth row. Upon further consideration, the possibility of top jaw to bottom jaw differences, and right side to left side, differences may still have some validity. Teeth in the top jaw are anchored in a fixed maxilla, whereas teeth in the lower jaw are attached to a free moving mandible. Does the process of lower teeth moving and occluding with fixed upper teeth have an effect on the microwear features formed on these teeth? Similarly, it may only seem logical that teeth on the right side will experience the same amount of abrasion as the left side, but it has been shown that most individuals have a dominant side on which they do most of their chewing. Is the dominance of one side reflected in their microwear pattern? The test for within individual differences in this study, for all the individuals with more than one tooth available, was intended to answer these questions.

For the phase II crushing/grinding facets used in this study, only one significant differences was found for any of the top to bottom, or right to left side comparisons. The exception to this finding was the significance found in the scratch density and average feature width of tooth 16 to 46 in individual 20. These isolated anomalies found in one pair of teeth in one individual cannot be easily explained, and may represent sampling error, or something undetectable at this time, such as a malocclusion in the individual. Due to the lack of sufficient information regarding the functional aspects of the occlusion and jaw mechanics for the individuals tested, any comments made regarding this anomaly would be pure speculation. It must also be recognize that the sample size used for these comparisons was small. Only eight pairs of teeth were used for top to bottom comparison, as well as for right to left comparisons. The lack of significant difference found between all the within individual comparisons may indicate a true homogeneity of the teeth, or it may represent an inability for the present study to detect a difference due to the selection of parameters used for comparison, experimental error, or a small sample size. A larger sample size with known functional parameters will be needed to verify these results.

Overall, the first molars of each individual in this study did not show any statistical differences in their microwear pattern. Therefore, all the first molars were pooled within each of the individuals to provide a larger sampling of that individual for statistical testing of between individual differences.

6.2.4. Between Individual Differences

Analysis of variance was used to look for differences between the nine individuals in this study. As the results have indicated, there were differences found between individuals for all the traditional parameters that involved feature count. The differences were all highly significant. Significant differences between individuals were found with the new parameters pit-area to scratch-area ratio and average length to width ratio. Since the analysis was conducted on an unknown sample, it is not possible at this time to determine which group of parameters was more accurate in its assessment of microwear pattern in relation to diet consumed, only that one group appears to be able to detect differences between individuals.

The analysis of variance for many of the parameters tested indicates a significant difference between the nine individuals. This statistical test does not, however, indicate whether some or all the individuals are different. Since the sample sizes are widely different for each individual (the number of teeth used for each individual vary from one to four), it was not possible to use their variances to detect mean differences between each individual. When the individual parameter values were plotted graphically, visual examination for individual differences that the parameters had detected was possible. All the bar graphs were displayed with the individuals in ranked order according to their average length to width ratio (Fig. 15). Displayed in this fashion, it was possible to assess the correlation of the other significant parameters in relation to average length to width ratio (Fig. 15a), the parameter that was potentially the most significant in detecting dietary differences. The parameter of pit to scratch ratio (Fig. 15g) and pit-area to scratch-area ratio (Fig. 15h) showed a similar pattern to average length to width ratio, although the individual ranked orders would be slightly different for these parameters.

As mentioned in the methods section, the ability to fully assess a microwear pattern depends on more than one parameter. Two or more parameters are required to quantify the size, shape, and number of features that make up a microwear pattern.

When two parameters were considered at the same time (Fig. 16), the separation between individuals become slightly clearer. Traditional pit to scratch ratio and feature density showed a clear separation of individuals 16 and 23 from the rest of the individuals. They appeared to be furthest separated from individuals 13 and 15 (Fig. 16c). The use of pit-area to scratch-area ratio and feature density shows a similar pattern, but the distinction of 16 and 23 from the rest of the individuals became less clear (Fig. 16d). With all the discussion regarding the findings of individual differences, one must be cognizant of the great deal of variation within individuals. This was due to the unbalanced sample size used for each individual, which resulted in our inability to conduct statistical tests for individual comparisons in the first place.

The plot of average feature length against average feature width compared both the average size and shape of features between individuals (Fig. 16a). Again the greatest differences were found between individual 13 and the two similar individuals, 16 and 23. The plot of average ratio against feature density brings the factor of numbers of features into consideration (Fig. 16b). This graph also clearly separates out the same individuals. In addition, individual 40 appeared to be significantly different from some of the others. When these two plots are examined together, the size, shape and number of features were all represented; the separation of the individuals resulting from the examination of these two plots have true differences in their microwear patterns. It appears that significant differences can only be shown between individuals at the two opposite ends of the parameter spectrum showing individual differences. The total number of individuals showing significant differences found with this comprehensive method was always greater than what each parameter can distinguish when used alone; thus showing the greater discriminating power of this new method.

When all the factors were considered in a microwear analysis, it is then possible to truly take full potential of the microwear pattern in dietary interpretation. That is not to say that all aspects of the microwear features must be involved in dietary interpretation, but without examining all these factors, one may reach an erroneous conclusion that one may not have made if all the factors had been considered. By employing an analytic method that include the parameters of average feature length, average feature width, and feature density, all the important factors that will identify a microwear pattern are included. The real test for significant factors in dietary interpretation can only be accomplished with a large sample of individuals with known diet.

6.2.5. Comparison to other studies

There has not been any significant baseline data collected for human samples. Therefore, the interpretation of diet for most human material has been extrapolated from animal studies, assisted by other methods of dietary determination. The main source that most resemble early humans would be other species from the primate order. Teaford and Walker (1984) completed a study of microwear patterns from seven species of primates with known diets. Three of the seven species (*C. albigena, C. apella, and P. pygmaeus*) were known hard fruit and nut feeders. Three others (*G. gorilla, A. palliata, and C guereza*) had a diet with relatively high proportions of leaves, stems, and flowers. The last species (*P. troglodytes*) was a known mixed feeder. The average length and width of the features were measured and compared (Fig. 18a). Significant differences were found between the three types of diets. The authors also included samples from several *S. indicus*, an extinct ramapithecine from the Miocene era. The diet of *S. indicus* was in dispute, and the microwear comparison to seven primates of known diet found them to be mixed feeders.

Similarly, the feature length and width data from the Ganj Dareh individuals were compared with the primate microwear data (Fig. 18a). The results show that Ganj Dareh individuals most resemble, and are in fact indistinguishable from *P.troglodytes*, a mixed feeder. To demonstrate that the plot of feature length vs. width has the ability to distinguish both shape and size, one just has to examine the position of the Ganj Dareh individuals on this graph. The Ganj Dareh individuals showed the shortest average length of features in the group but average width, which demonstrated the shape of the features. The average size of the features can be determined by their distance from the intercept of the two axes. As one moves further away from the intercept of the axes, the feature size increases. Ganj Dareh individuals also showed the smallest size features of the group.

Teaford and Walker, at that time, believed that it was necessary to distinguish between pits and scratches. They arbitrarily chose the length to width ratio of 10:1 as a cut off point to distinguish pits from scratches. The percentage of pits of each species, and that of the Ganj Dareh samples, are shown in Figure 18c. The use of pits and scratches to assess the microwear patterns of these primates, with an arbitrary length to width ratio of 10 to define pits and scratches, changed the ranking of Ganj Dareh individuals, to a position that was most similar to *P. pygmaeus and C. apella*, both hard object feeders. This was not the only inconsistent finding. The authors reported that the ranking of *A. palliata* also changed when different length to width ratios for defining pits and scratches were used. The authors did not publish the data of pits to scratch ratio other than for the ratio of 10, therefore this study was unable to assess the kind of changes it would have made for Ganj Dareh samples in relation to the rest of the species. It was more important to note the fact that the arbitrarily chosen cut off points for pits and scratches will have an affect on the results of microwear analysis. Due to the inconsistent way that this ratio was used, it should be replaced by the parameter average length to width ratio. The average length to width ratio for each species are shown, in graphic form, in Figure 18b. Both the Ganj Dareh individuals and the *A. palliata* samples show a difference in their ranking against the rest of the species with this non-categorical parameter.

Molleson *et al.* (1993) reported a microwear study involving Mesolithic (1a) and Neolithic (2A and 2B) Abu Hureyra individuals, primarily meat eating modern individuals from Spitalfield, and modern individuals from Abu Hureyra (2C) with a mixed diet. The data for proportional area of pits vs feature density for all the samples are plotted and illustrated in Figure 17. The Ganj Dareh samples in this study were contemporaries with the Neolithic Ib Abu Hureyra individuals, and both groups were found in the Near East. The Ganj Dareh data have been included for comparison. From the examination of this graph, the Ganj Dareh individuals most certainly grouped with the modern individuals that consumed processed and cooked foods. Unfortunately no standard deviations were presented with Molleson's result, making it impossible to determine a significant difference between any of the individuals.

The results that suggested Ganj Dareh individuals having had a similar diet to the modern samples instead of those individuals found in early Neolithic (Neolithic Ib) times is very surprising. This would tend to imply a greater ability of the Ganj Dareh individuals to process and cook their food than the archaeological findings would suggest. The Mesolithic Abu Hureyra individuals appeared quite close to the Ganj Dareh and modern individuals in this graph as well. There was a possibility that all three groups of individuals were statistically indistinguishable, should data be available for such tests. If such was the case, Ganj Dareh individuals in this study may represent a population that was still in a stage of dietary transition that was more closely related to a Mesolithic form of subsistence. Neolithic Ib population was found to have domesticated wheat, and had included wheat in their diet. Domesticated grains generally have larger grain size, together with small stone inclusions from the grinding stones used in their preparation, may have contributed to a much coarser diet than the Mesolithic diet. Ganj Dareh population did not have domesticated grains, with the exception of barley. The barley found at the site may not have been consumed directly, but may have been used primarily for brewing of beer. If such was the case, then Ganj Dareh individuals would still have relied on the hunter-gatherer's way of subsistence, with domesticated barley playing a minor or insignificant role in their diet and microwear formation. The different scenarios suggested here cannot be substantiated without further statistical testing and archaeological evidence.

The major criticism for this comparison, and possible explanation for the discrepancy in results, must be the different methods used in the microwear analysis. Molleson utilized a low magnification (180X) SEM photomicrograph for her analysis. As discussed in the literature review, the ability to identify and measure features at low magnification has been subjected to criticism. At the same time, when higher magnifications are used, the field area becomes exponentially smaller, increasing the chance of sampling error, and often truncating larger features that could be of some significance. The general opinion of the research field is that microwear studies conducted at different magnifications may be measuring completely different sets of features; the data recorded are for features that are most apparent at that specific level of magnification. Therefore, the difference in magnification used for microwear analysis between these two studies may have been significant enough to render the studies incomparable. Another factor that may have contributed to this result is the fact that very small number of samples were used for each group. As can be seen from Figure 17, the samples that represent each group consisted of between 3 and 5 individuals. Finally, Molleson's study indicated no significant differences between any of the groups when the traditional pit to scratch ratio was used for comparison. The non-significant differences found between individuals shown by their pit to scratch ratio may have been due to the small sample size used, and prompted the search for a new parameter that can separate the groups. The new parameter "area devoted to pits" was introduced as the solution. The data for the new parameter "area devoted to pits" was obtained by averaging the results of a sampling of twenty pits. The use of a sample within a sample to represent the individual, together with the difficulties of accurately measuring feature width at low magnification, may have created a high level of sampling error that was not discussed in the study.

The controversial results from the comparison of Molleson's human samples and the present Ganj Dareh samples may, however, indicate a true difference between the Ganj Dareh and Abu Hureyra early Neolithic samples, or merely reflect a difference in analytic methods. Resolution of this controversial comparison awaits results from standardized testing for both samples.

The Ganj Dareh group were from a population that demonstrated certain innovations towards control of their dietary resources and preparation methods in their food. The presence of domesticated goats and barley suggest at least a partial control over their dietary sources. The use of tools for food preparation was demonstrated by the findings of pestles, mortars and rubbing stones. Kilns used for firing of ceramics suggested the possibility of cooking and storing of food. The ability to boil food in a large vessel can dramatically reduce the hardness of food. The Ganj Dareh samples that was analyzed in this study certainly had all of the above attributes, with the exception of ceramic pots. The presence of kilns and cooking vessels was found in post level D in the excavation site. It is uncertain as to the availability of these utensils to the individuals in level D, a population from which majority of the individuals in this study were sampled.

The significance of the comparisons between Ganj Dareh and those from different species of primates and other human samples was that they may assist in determining the mode of subsistence of the Ganj Dareh population. Since Tepe Ganj Dareh represents one of the earliest sites of a sedentary way of life, the change in hunter-gatherer to agricultural subsistence may have been accompanied by a significant change in dietary pattern. The suggestion that these individuals may have been mixed feeders, and that they may have consumed processed and cooked foods tend to agree with archaeological evidence of this site, but at a slightly later date than the level of excavation where these individual were discovered would suggest. The majority of individuals were found in level D of the excavation; but evidence for the technical sophistication in processing and cooking soft foods were not found until post level D. The health status of Ganj Dareh individuals shows a pattern of low disease, and high mortality rate, suggesting close proximity in time to hunter-gatherer way of life. Since the dental microwear analysis did not match with such a mode of subsistence, this would indicate that the change in subsistence must have occurred relatively recently. These apparent discrepancies cannot be resolved until evidence from comparisons of the microwear patterns to other standardized human data can be obtained, or further archaeological evidence is discovered.

6.3 Modern samples

The microwear patterns of the two modern samples, when examined visually, were dramatically different than those found on the prehistoric samples used in this study. As expected in modern population, tooth wear in our samples appear to be very minimal. The first molars were examined in our sample, and at the gross level, facets were found only on the cusp tips. Even when examined with the SEM, crushing/grinding (phase II) facets are rarely found, and relatively few microwear features were found in the areas where phase II facets should be.

In general, photomicrographs were taken wherever microwear features could be found. In these samples, most of the features were found on cusp tip facets. As the results indicate, the features found were mostly large, irregular pits, and scratches with random orientation (Fig. 17). The facets were not covered with fine striations that would indicate significant tooth to tooth contact, or attritional wear. The pits often out number the scratches. This was a surprising find, since this type of microwear feature would normally suggest a highly abrasive and destructive diet, with an accompanying high turnover rate. This suggestion would be contrary to the evidently general lack of wear at the gross level, and what one might expect of a modern, highly processed diet.

The answer to this puzzling find may be revealed as we further examine the microwear turnover in our longitudinal results. As we can see from the comparison of initial and final photomicrographs (Fig. 21), very few new features were found on these surfaces. Teaford and Tylenda (1991) had found that the rate of microwear turnover was approximately 60 days, in a sample of nine healthy adults. The time between the initial and final impressions for Teaford's study was never more than 7 days. Although formal analysis of the rate of turnover was not done for this sample, due to the small sample size, the rate of microwear formation in this sample appears to be less than Teaford's sample. Subject A demonstrated a rate of microwear turnover of 238 days. Subject B demonstrated a rate of 707 days to replace the 177 features found initially.

With such a slow rate of feature turnover, one would suspect that their diets were very non-abrasive. With the fact that all of the new features found were of much smaller size than the large pits or gouges that are evident when one first examined the photomicrographs, one would suspect that the large pits and gouges are due to rare catastrophic events that are overshadowing the underlying normal functional wear. The creation of such features may represent occasions where the individual used their teeth as a tool, parafunctional activities, or when they unsuspectingly occluded on abrasive inclusions in their otherwise very soft meal. These large features would require significantly greater time to be reworked and erased. Perhaps they can only be replaced by similarly large features formed during the next catastrophic event. The microwear features formed during the slow process of functional wear was superimposed on this dramatic microwear pattern, and would take an extremely long time before this functional wear could alter the total microwear pattern. The occurrence of such catastrophic events may be highly unpredictable, and may vary greatly between individuals, making the rate of turnover very hard to predict.

If the majority of these features were indeed the work of catastrophic events that were not related to normal functional wear, the analysis of the microwear pattern would not in any way be a good predictor of dietary content. In the subjects used for this study, both of them confessed to occasional parafunctional activity and/or dysfunction. The act of clenching and bruxing may create microwear patterns that have not been analyzed before. The lack of a sufficiently large sample precludes detailed analysis and discussion of these speculations. A larger sample size with a control sample and longer and more repeated follow-up impressions would be needed to observe this phenomenon.

7.0 CONCLUSIONS

The present study has undertaken methodological and technical steps to reduce the amount of time required for completing a microwear analysis, at the same time providing a comprehensive standardized microwear analysis method that will give accurate results for future comparisons between different studies. The results indicated that in order to account for all the characteristics of a microwear pattern, the method used in its evaluation must include evaluation of the size, shape and number of the features present. A new method of microwear analysis, including parameters of average feature length, average feature width, and feature density, has the ability to meet this requirement, and has been shown to possess the best discriminatory ability when used with the Ganj Dareh sample.

The use of a semi-automated system in this study has allowed, for the first time, the combination of a highly accurate measurement of feature dimensions, and efficiency. The search for a fully automated system remains a priority, as the efficiency of data collection and analysis continues to be the time and effort limiting factor in dental microwear analyses.

These improvements in the microwear analysis have been applied to the Neolithic Ganj Dareh human samples in an effort to determine their diet. The results of the analysis indicate that differences can be found between individuals of this group. The lack of a balanced sample prevented complete statistical analysis to detect specific differences between each of the individuals.

The microwear patterns of these individuals were also compared with that of seven species of primate, and of Mesolithic, Neolithic, and modern human samples obtained from other studies. The comparison to other primates indicated that the Ganj Dareh individuals were mixed hard and soft object feeders, while comparisons to other human samples indicate that the Ganj Dareh samples had microwear patterns that most resembled modern human population with soft processed diets. This apparent discrepancy in results was best explained by the fact that all the studies utilized differing methods to obtain their results, again emphasizing the importance of a standardized method to allow direct comparison of different microwear studies. The inadequacies of these studies have prevented accurate comparisons of results; the problems encountered and solutions required have been discussed.

Qualitative microwear analyses of two living individuals have been conducted. The objective was to assess the potential of utilizing microwear analysis for dietary and functional discrimination in modern individuals. The results of the qualitative analysis indicated that the microwear pattern found in these individuals, and perhaps in the modern population may not indicate dietary differences, but in fact indicate parafunctional activities. The search for inferences to these microwear patterns require the efforts in future studies.

7.1 Future considerations

The present study has attempted to develop an automated system for microwear analysis. Although the effort has only been partially successful, the potential and limitations for developing such a system has been discussed. The problem of feature discrimination by an automated system has been the major stumbling block to analysis automation, and future efforts should concentrate on solving this problem. Traditionally, microwear analysis has utilized two dimensional images of the tooth surface for feature identification. Features represented by a two dimensional image can only be identified by their differences in shade, that is contrasted against a different background shade. As discussed previously, biological variation makes automated feature identification through shade differences impractical. In order to assist feature identification, three dimensional imaging appears to have the greatest potential. Features can be located through a change in depth from the surrounding surface with a three dimensional image of the total surface. SEM approaches have the advantage of high magnification and large depth of field, but are limited by their inability to create three dimensional images. Stereoscopic views of the SEM images are not true three dimensional images; they just provide different views of an image that when viewed together, suggest a three dimensional interpretation. The most promising method of obtaining three dimensional images of the surface appears to be with the use of surface laser scanning. As mentioned in the literature review, laser scanning has been employed for three dimensional scanning of tooth surfaces, but it has not been attempted for producing high resolution images that microwear analysis requires. The efficiency and practicality of dental microwear analysis requires the development of such an automated system.

A comprehensive method of microwear analysis has been introduced in this study. Unfortunately due to the limitation of available samples, the accuracy and significance of this method cannot be fully demonstrated. The parameters chosen for this new method of analysis have proven to have a greater discriminating power than any of the traditional parameters when used separately. The limitations of the present sample prevented our ability to correlate the significant differences found to dietary differences. The use of this comprehensive method with a large balanced sample of individuals with known diets would reveal the true potential of this method, and provide baseline data against which future work can be compared. Additional testing with other human samples found in similar regions and time periods would then place the present sample in context with their contemporaries. Along with other archaeological findings, microwear analysis can then be used for interpretations other than diet, such as technical advances in food storage, gathering, and preparation.

The results from the qualitative analysis of living humans indicated a peculiar but significant pattern of microwear features. The tentative conclusion reached was that the microwear patterns found were not indicative of normal functional wear, but were probably due to parafunctional events; in which case, dietary interpretation would not be possible with microwear analysis of modern humans. The pattern of microwear does suggest the need for future efforts to isolate a cause for their formation. The most likely candidate may be parafunctional activities related to stress or biological dysfunction. Microwear patterns resulting from tooth contacts in relation to clenching and bruxing have not been thoroughly investigated. The extremely low rate of microwear turnover found in this study agrees with the overall gross wearing of the teeth but contradicts the microwear pattern found. The microwear rate of turnover in industrialized populations must be further investigated with a larger sample size and detailed functional and dietary information available for the individuals.

The use of microwear turnover to determine actual amount of tooth wear has the advantage of determining amount of tooth wear in a relatively short time. Microwear features have been shown to be formed on a daily basis. Significant number of new features can be found on impressions of teeth taken days or weeks apart. Since gross wearing of teeth is extremely slow, the rapid determination of tooth wear can have a tremendous impact in dental research that require an estimation of tooth wear. Development of a reliable method for determining the rate of microwear turnover in relation to the amount of enamel loss will be invaluable in many areas of dental research.

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