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Human Dental Microwear Caused by Calcium Oxalate Phytoliths in Prehistoric Diet of the Lower Pecos Region, Texas

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ABSTRACT Recent research demonstrates that silica phytoliths of dietary origin are associated with microwear of human teeth. Previous research has shown that severe enamel microwear and dental wear characterizes Archaic hunter-gatherers in the lower Pecos region of west Texas. Calcium oxalate crystals are especially common in Archaic coprolites. The vast majority are derived from prickly pear and agave, which were the dietary staples in west Texas for 6,000 years. The calcium oxalate phytoliths are harder than enamel. Therefore, calcium oxalate crystals are the most likely source of previously documented dental microwear and wear in the lower Pecos region. *Am J Phys Anthropol* 107:297-304, 1998. © 1998 Wiley-Liss, Inc.

Previous research shows that the hunter-gatherers of the lower Pecos region had especially pronounced dental pathology. Dental wear, dental caries, and premortem tooth loss were the most consistent health problems faced by ancient Texas hunter-gatherers (Hartnady, 1986; Hartnady and Rose, 1991; Marks et al., 1985, 1988; Reinhard et al., 1989; Turpin et al., 1986). Stable carbon isotopic study of mummies and skeletons (Huebner, 1991) and botanical study of coprolites (Reinhard, 1992) shows that these hunter-gatherers were reliant on desert succulent plants, especially prickly pear and agave. These plants have been found to have a high concentration of calcium oxalate phytoliths (Danielson, 1993).

Dentition from the lower Pecos has been particularly well studied using SEM by previous researchers (Hartnady and Rose, 1991; Marks et al., 1985, 1988) and also by macroscopic study (Hartnady, 1986; Turpin et al., 1986). An analysis of adult dentitions from the Seminole Sink site showed extreme dental wear and dentin exposure (Turpin et al., 1986; Marks et al., 1985, 1988). Of the ten adult mandibles and maxillae, all are edentulous or nearly edentulous. Of the molars

that remained in the alveolar bone, 70% had smooth, polished occlusal surfaces with rounded occlusal margins. Anterior teeth had no or little enamel left with extensive dentin exposure. Deciduous teeth from individuals ranging in age at death between 2-5 years exhibited loss of enamel on the occlusal surfaces and dentin exposure. From the same site, 64 loose teeth were recovered. SEM analyses of these teeth revealed that the polished enamel surfaces had microwear in the form of gouges, striations, and pits. Striations and groups were distinguished by width (Turpin et al., 1986; Marks et al., 1985, 1988). Striation widths ranged between 1.2 and 4.7 μms , gouge widths ranged between 0.3 to 2.8 μms . Pits caused by compression ranged between 1.1 and 3.8 μms . Hartnady completed a study of 52 individuals from the lower Pecos spanning the full temporal range of the Archaic period (8000 BC to AD 1000). This study revealed that by age 25, essentially all molars were

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lost and by age 40, all adults were virtually edentulous. Turpin et al. (1986) found the same pattern in a study of Archaic mummies. In fact, dental wear and tooth loss was a much more significant problem for Archaic peoples in the lower Pecos relative to those of the Texas coast, coastal plains, or Edwards Plateau regions (Reinhard et al., 1989).

Several dietary explanations for the wear have been postulated. These include: 1) a diet high in grinding stone grit (Hartnady and Rose, 1991), 2) a diet high in plant fiber (Turpin et al., 1986), and 3) dietary dependence on hard seeds (Turpin et al., 1986). We are not content with these explanations for the following reasons. First, although Teaford and Lytle (1996) present particularly compelling documentation that grit from grinding stones is a significant source of dental wear, analysis of coprolites from the lower Pecos by many researchers does not reveal that ground food was a major part of the diet (Bryant, 1974a,b, 1986; Bryant and Larson, 1968; Bryant and Williams-Dean, 1975; Edwards, 1990; Reinhard, 1992; Sobolik, 1988, 1991, 1994; Stock, 1983; Williams-Dean, 1978, 1984). Reinhard examined microscopic residue from 96 lower Pecos coprolites and found no grinding stone grit. Grit from grinding stones can be found in coprolites in the form of small rounded mineral fragments. Second, although plant fiber is very common in coprolites, cellulose alone is not hard enough to produce striations on enamel. Finally, although hard seeds in the lower Pecos diet have been implicated as a cause of "microfractures" in dentin after the enamel was abraded away (Hartnady and Rose, 1991), the seeds have not been implicated as a cause of enamel striations.

We feel that there must be an alternative explanation for the dental wear. Because there is a rich record of diet from coprolite and stable isotopic study for the lower Pecos, we believe that the cause of the wear is evident in those dietary evaluations. Phytoliths are ubiquitous in lower Pecos coprolites, as demonstrated independently by several researchers (Bryant, 1974a,b, 1986; Bryant and Williams-Dean, 1975; Edwards, 1990; Reinhard, 1992; Sobolik, 1988). Fox et al. (1996) present an illuminating study on

the role of dietary silica phytoliths in the causation of microwear on enamel surfaces of human teeth. They clearly document that silica phytoliths of dietary origin caused microwear. In coprolite studies of Archaic hunter-gatherer cultures from southwest North America, phytoliths have been observed as a significant coprolite component (for review, see Reinhard and Bryant, 1992). However, the phytoliths that are most common are not silica phytoliths. Instead, calcium oxalate phytoliths are present in Archaic coprolites. Because calcium oxalate phytoliths are perceived to have less taxonomic value than silica phytoliths, they are less often studied. Their role in dental wear has gone unevaluated.

In the light of Fox et al. (1996), we hypothesize that calcium oxalate phytoliths were the cause of the lower Pecos enamel wear. If phytoliths did cause dental wear, we expect that they will be abundant in coprolites and harder than enamel. Below, we present our analysis of the phytolith content of lower Pecos coprolites and an assessment of the potential of calcium oxalate phytoliths in dental microwear and wear. If the phytolith content of coprolites was high and if the identified chemical structure could be demonstrated to be harder than enamel, then phytoliths can be implicated as the cause of previously described enamel wear among prehistoric lower Pecos hunter-gatherers.

MATERIALS AND METHODS

Modern comparative plant materials

Published descriptions, especially for grasses, were used for identifying some phytoliths. However, since calcium oxalate phytoliths are often undescribed, with one notable exception (Jones and Bryant, 1992), it became necessary to visit the lower Pecos to collect modern plants to establish a comparative collection. A phytolith reference collection was made by collecting wild plants in the lower Pecos and extracting phytoliths from them. These included the major plant foods recovered from coprolites (agave, yucca, sotol, prickly pear, wild onion, mesquite), and other wild plant foods and medicinal herbs that occur in the area (goosefoot, pigweed, fourwing saltbush, jimsonweed, rice grass, acacia, river cane, bean, gourd, and

horsetail). The plant parts that were known to be eaten from agave, sotol, yucca, and prickly pear were selected for extraction.

Phytoliths were extracted using the following protocol. Plant samples were washed with distilled water to remove extraneous soil and the samples were finely chopped. The chopped plants were softened in household Clorox (5.25% sodium hypochlorite) for several days until visual observation showed that the tissue was oxidized and phytoliths were liberated. Floating organic remains were removed and the fluid was poured into centrifuge tubes. The phytoliths were concentrated by centrifugation and formed a solid plug in the bottom of the centrifuge tubes beneath the fluid supernatant. The supernatant was poured off, leaving the phytolith plug in the tube. Next, 95% ethyl alcohol was added to each tube and the phytoliths were stirred back into solution (a vortex stirrer works well for this process). The phytoliths and alcohol were transferred to screw-cap vials. The screw-cap vials are not air-tight; therefore, the alcohol will evaporate even if parafilm is used to seal the vials. Therefore, the fluid levels in the vials need to be monitored on a regular basis.

Analyzing phytoliths from coprolites

Reinhard (1992) examined 96 coprolites for dietary and parasitological analysis. Coprolite residues from these coprolites were reexamined to test for phytolith content. Fourteen coprolites from Hinds cave in the lower Pecos dating between 500 BC and AD 1000 were selected for more detailed analysis. They were processed in 30% hydrogen peroxide and potassium dichromate ($K_2Cr_2O_7$). This technique was originally applied by Jones (1988) and was refined for application to coprolites by Danielson (1993).

The hydrogen peroxide and potassium dichromate procedure is as follows: One gram of coprolite (40 mls in volume) was cut from the interior of the coprolite and gently crushed with a precleaned mortar and pestle. The interior of the coprolite was used to reduce contamination with non-fecal soil matrix. The crushed sample was placed in a clean 600 ml beaker. In a fume hood, just enough 30% hydrogen peroxide was added to cover the material. The beaker was swirled

by hand until all material was covered with peroxide and then was left to stand for 5 min. Approximately 0.25 grams of potassium dichromate crystals were added over the crushed coprolite material and the beaker was swirled to aid in the dispersion of the chemical throughout the sample. A delayed, rather violent bubbling resulted. (To prevent the sample from bubbling over, 95% ethyl alcohol was sprayed over the sample.) When the reaction began to abate, approximately 1–2 ml of hydrogen peroxide was added to renew the oxidizing process. When all or most of the organic material was digested, the sample stood until the reaction ceased and the beaker cooled. The contents of the beaker were poured into 12 ml glass test tubes and concentrated by centrifugation. The remains were rinsed with distilled water and centrifuged three times to eliminate any hydrogen peroxide and potassium dichromate residues. After decanting the last water wash, approximately 8 ml of 95% ethyl alcohol were added and the phytoliths were concentrated by centrifugation. The samples were transferred to 1 dram vials for storage in 95% ethyl alcohol in screw-cap vials sealed with parafilm at room temperature. The reader should be aware that this process is extremely dangerous and difficult to control. Double gloves, lab coat, closed shoes, apron, and goggles must be worn. The process must be conducted in a fume hood.

The phytoliths were then identified taxonomically by comparison to the reference samples with a compound light microscope at 400x and 1000x magnification. The relative abundance of different phytoliths was evaluated based on 200 phytolith counts for each coprolite sample. The 200 item count has become the standard for microfossil analysis based on Barkley's (1954) demonstration that a count of 200 items is essential for statistical representation of microfossils. Pollen was the study medium in Barkley's case, but Pearsall (1989) describes the utility of 200 counts for phytolith studies.

Quantifying phytolith content of coprolites

A plastic 12 ml graduated centrifuge tube marked at 1-ml intervals was used to quantify the coprolite residue. The phytoliths

TABLE 1. Phytolith counts from 14 coprolites from Hinds Cave Texas

Sample #	Agavaceae	Cactaceae	Choridoid	Festucoid	Panicoid	Fabaceae
1	198	5	0	0	1	0
2	198	2	0	0	0	0
3	102	103	0	0	0	0
5	198	2	0	0	0	0
7	188	12	0	0	0	0
8	200	0	0	0	0	0
10	188	10	0	0	0	2
14	163	73	0	0	0	0
15	197	6	0	2	0	0
16	160	40	0	0	0	0
17	201	25	0	0	0	0
18	180	20	0	0	0	0
19	188	4	2	0	4	1
25	152	48	0	0	0	0

Choridoid, Festucoid, and Panicoid refer to three taxa of grasses that produce distinct phytoliths. Agavaceae and Cactaceae phytoliths are calcium oxalate. The various grass phytoliths are silica-based. The data show that over 99% of the counted phytoliths are calcium-based.

were concentrated in each tube by centrifugation. The amount of solid phytolith plug was then measured by comparison with the ml marks on the tube. The volume of coprolite processed was 40 ml. Therefore, the amount of phytoliths in each coprolite was determined by the ratio of phytoliths in the tube to 40 ml of coprolites. For example, if 10 ml of phytoliths were recovered, this represents 25% of the original 40 ml volume of coprolite.

Determining chemical composition. An x-ray microprobe was used to determine the chemical composition of the phytoliths. The specific instrument used was a Cambridge SEM with attached Keveex analyzer. Working distance was 7–10 mm, voltage at 5,000 kv, with variable spot size.

Testing phytolith hardness. To test phytolith hardness relative to enamel, a Moh test was used. The Moh test evaluates hardness of materials by the ability of test materials to scratch the surface of apatite tiles of known hardness. The Moh scale ranges from 1–10 with 1 being the softest. The hardness of enamel has been demonstrated to be between 4.5 to 5.0 by previous research (Baker et al. 1959). Therefore, the phytoliths were tested against the 4.5 and 5.0 tiles.

Grinding stone grit. Grinding stone grit is a documented source of microwear (Teaford and Lytle, 1996). As mentioned above, ground food is not a significant part of the diet. However, we examined the coprolite residues for evidence of grit from grinding

stones in the form of nonphytolith mineral fragments.

RESULTS

Comparison of the phytoliths extracted from the coprolites with those from modern samples shows that plants in the Agavaceae (agave) and the Cactaceae (in this case, consistent with prickly pear, *Opuntia* ssp.) were overwhelmingly represented in the coprolites (Table 1). Both plants are CAM desert succulents. CAM plants are those that can produce either C3 or C4 signals, depending on environmental conditions. In the lower Pecos, CAM plants produce C4-like stable carbon isotope ratios. All samples contained sufficient phytoliths to obtain 200 phytolith counts. All 96 coprolites contained phytoliths from prickly pear and/or plants in the agave family.

The detailed analysis of 14 coprolites was insightful. All coprolites contained an abundance of phytoliths. The volumes of phytoliths ranged between 4 ml to 8 ml. Considering that 40 ml of coprolite were processed for each sample, this shows that one-tenth to two-tenths of the volume of a coprolite is typically composed of phytoliths.

The chemical composition of the Agavaceae and Cactaceae phytoliths is dominated by calcium. Since the main calcium-based phytoliths described are of calcium oxalate, we assume that these are calcium oxalate crystals (Fig. 1).

The calcium oxalate phytoliths are harder than enamel. In the Moh test, they abraded test tiles at 4.5 and 5.0 on the Moh scale.

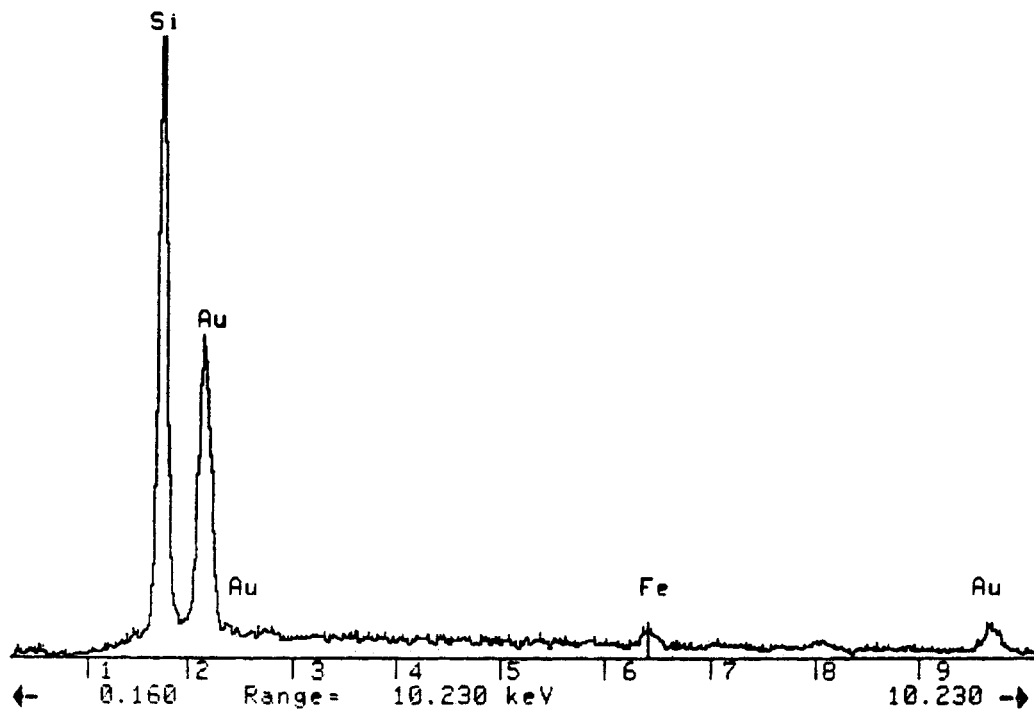
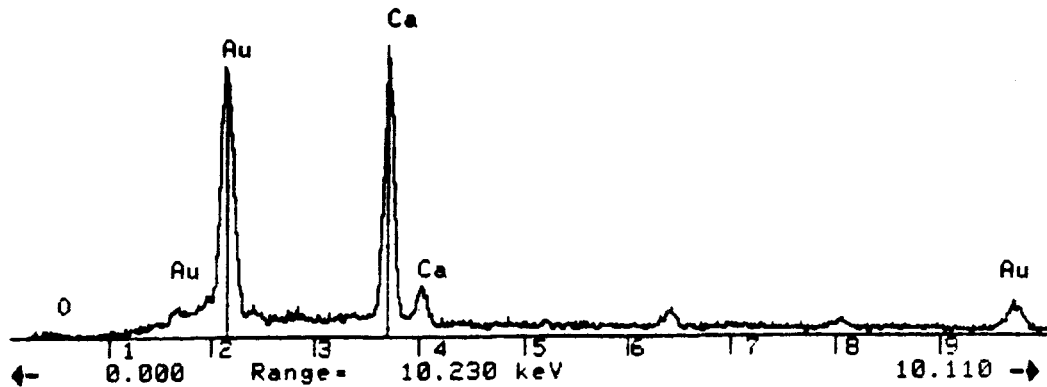


Fig. 1. Phytoliths are of two distinct chemical structures: calcium oxalate and silica. The upper spectrum is from a phytolith from *Agave* and is dominated by calcium. The lower spectrum is from an *Equisetum* (horsetail) phytolith and is dominated by silica. The phytoliths from the Hinds Cave coprolites are consistent with the upper spectrum.

DISCUSSION

Our hypothesis that calcium oxalate phytoliths caused dental microwear and wear in the lower Pecos is supported by our analysis. The coprolites contain substantial amounts of phytoliths. These are morphologically com-

parable to phytoliths in modern desert succulents of the area. The chemistry of the phytoliths is consistent with calcium oxalate phytoliths. These crystals have points and edges (see below) that are similar in dimension to the scratches measured by SEM

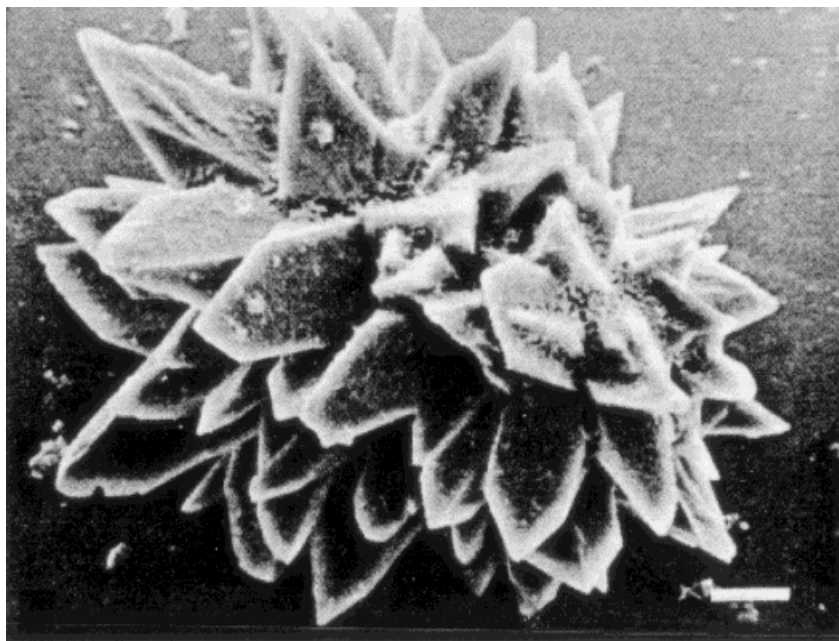


Fig. 2. This scanning electron micrograph of a prickly pear phytolith from a coprolite shows the morphology typical of this type of cactus (Jones and Bryant, 1992). The sharp structures that protrude from all angles of the phytolith would be very destructive to chewing surfaces. Bar = 5 μ m.

study of prehistoric teeth by Marks et al. (1985). The calcium oxalate phytoliths are hard enough to abrade enamel, as shown by the Moh test. In the authors' experience, there is no other dietary constituent that is as prevalent, nor more abrasive, than the calcium oxalate phytoliths that have been found in lower Pecos coprolites. These have long been noted as coprolite constituents (Bryant and Williams-Dean, 1975), but they have never before been tested as a source of dental wear.

Our phytolith findings are consistent with stable carbon isotopic analysis of the lower Pecos which shows a reliance on these desert succulents (Huebner, 1991). Late Archaic skeletons analyzed by Huebner had $\delta^{13}\text{C}$ collagen values that range from -15.7 to -12.6. Huebner demonstrates that these values are consistent with CAM plants. This is also consistent with 30 years of coprolite analyses by various researchers, which consistently show a dietary reliance on cactus and agave families (Bryant, 1974a,b, 1986; Bryant and Larsen, 1968; Bryant and Williams-Dean, 1975; Edwards, 1990; Rein-

hard, 1992; Sobolik, 1988, 1991, 1994; Stock, 1983; Williams-Dean, 1978, 1984). Importantly, Danielson (1993) demonstrated that these succulents are a major source of phytoliths.

It is important to emphasize that the phytoliths are free in the intestinal lumen. In microscopic analysis of 96 lower Pecos coprolites, Reinhard found that individual phytoliths predominate in material that is simply screened from coprolites without chemical treatment. This indicates that mastication liberated the phytoliths from plant tissue, and that the teeth were exposed to the phytoliths. This can be demonstrated by experimental chewing of phytolith-rich plant material. Teaford and Lytle (1996) provide a model for such research.

We suggest that cactus phytolith morphology is especially conducive to microwear of enamel. A thorough analysis of cactus phytolith morphology has been presented (Jones and Bryant, 1992). Cactus phytoliths range in size from 50 to 500 μm in diameter. Cactus calcium oxalate phytoliths are spheroidal with long projecting spikes (Fig. 2).

Intuitively, this morphology would be consistent with the objects that created microwear striations noted previously in lower Pecos teeth. This could also be evaluated by experimental mastication of cactus pads. Following the experimental design of Teaford and Lytle (1996), the relative effects of different plant taxa on enamel microwear should be evaluated.

Our findings contrast with some previous interpretations. Hartnady and Rose (1991) suggested that enamel striations were caused by dietary grit from grinding stones, dirt, and ash added to the foods. It is important to stress that we did not encounter grit from these sources in this analysis. Phytoliths are the sole source of abrasives as represented by coprolite studies.

Hartnady and Rose (1991) also propose that quid chewing was a source of microwear. Quids are fiber masses from plant foliage that are spit out after chewing. Typically, Archaic people in the lower Pecos chewed the leaf bases of cooked agave plants. Therefore, the quids are usually derived from agave in this area. Our findings are consistent with Hartnady and Rose's interpretation, but in a slightly different way. Hartnady and Rose argue that the fiber was a source of wear. We suggest that the phytoliths in the fiber matrix were the source of wear. This is especially true of plants in the Agavaceae, which have phytolith-rich vascular tissue. Plants in the Agavaceae were chewed extensively and quids of these plants are the most common finds in lower Pecos caves. In contrast, cactus phytoliths occur predominantly in the epidermis and cactus were not chewed as extensively as agave family species.

The reliance on desert succulents is not restricted to the lower Pecos. Reinhard (1992) presents a case that prickly pear and agave or yucca were widely eaten in the Great Basin, Mojave Desert, Colorado Plateau, and Sonoran Desert. Therefore, the effects of phytoliths on dental wear may also be evident among Archaic dentitions in these regions. This brings up one major point: the introduction of abrasives into diet prior to agriculture occurred with the consumption of phytolith-rich desert succulents. Depending on environment, abrasives were in the

prehistoric diet long before stone grinding became commonplace. Therefore, ancient dietary abrasives came from the foods themselves in the form of phytoliths, as opposed to introduced abrasives from food preparation tools such as grinding stones or from ash added to foods.

SUMMARY AND CONCLUSION

This study documents three important aspects of dietary abrasives in the lower Pecos of west Texas. First, dietary abrasives in the form of calcium oxalate phytoliths are very common and abundant in coprolites. Second, the phytoliths are derived from the main dietary staples in the region: prickly pear and agave. Third, the calcium oxalate crystals are harder than tooth enamel. This implies that the prehistoric dietary dependency on phytolith-rich desert succulent plants led to tooth wear which is the cause of abscesses and early tooth loss which are the most common forms of paleopathology in the region. The study has implications that extend beyond west Texas. Since cactus, agave, and other phytolith-rich plants were the dietary staples for much of the Archaic period in the desert West of North America, it is very likely that calcium oxalate crystals in other desert regions will be linked to prehistoric dental wear.

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