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Plant Microfossils Recovered from Dental Calculus at Casas Grandes, Mexico

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Craft Beer, Corn Smut, and Casas Grandes

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INTRODUCTION

In an attempt to better understand the diet and nutrition of the people of prehistoric Casas Grandes, we analyzed samples of dental calculus (tartar) recovered from the human remains of 110 people found at Paquimé and other sites in the Casas Grandes river valley in Chihuahua, Mexico. These remains are curated at the Museo de las Culturas del Norte in Casas Grandes and represent those who lived in the northwestern region of Chihuahua from about 700-1450 A.D. The samples were extracted by Kyle Waller during the summer of 2014, and Daniel King processed the samples for slide mounting later that year. Chad Yost, a PhD. candidate at the University of Arizona, performed the microscopy of the samples and provided a report of his findings in 2015.

Our research is based on previous projects in which dental calculus has been successfully used to help reconstruct prehistoric diets. As Boyadjian et al. (2007) suggest, analyzing plant remains in the form of microfossils "could render information about specific plants eaten and manipulated with the teeth. It is an ideal complementary method to stable isotopic reconstruction, because the dental calculus provides information about foods eaten a short time before death (from days to weeks, depending on the size of the deposit), whereas isotopic analysis provides a long-term perspective on diet."

METHODS

Dental calculus was removed from archaeological teeth by Kyle Waller at the Museo de las Culturas del Norte in June 2014 during analysis of the human skeletal remains. All individuals with teeth from the Convento site (Viejo period, 700-1200 A.D.) and Paquimé (Medio period, 1200-1450 A.D.) were examined macroscopically for adhering calculus. If calculus was noted, the tooth was cleaned with double-distilled water using a soft-bristled toothbrush. This was particularly necessary for Viejo period skeletons that appeared to have not been as sufficiently cleaned or organized as Medio period skeletons. The teeth were then allowed to air dry. A scalpel that had been sterilized in ethanol and rinsed in double-distilled water was used to remove the calculus samples from teeth, and the samples were then placed on weighing paper. Each calculus sample was placed in a labelled, sterile plastic sample vial, and weighed using a high precision milligram scale.

Once the calculus was removed, it had to be digested in order to extract the microfossils in preparation for slide creation and eventual microscopic analysis. Hydrochloric acid was first used to begin the digestion process, with little to no added heat or agitation. Second, the samples were rinsed using distilled water to stop further digestion. Each sample was processed in a mini centrifuge at 800 rpm for 20 seconds. Third, and finally, the samples were stored in ethylalcohol so as to prevent osmosis of any microfossil cells, primarily starches.

The samples were then mounted on microscope slides and microscopy was conducted with an Olympus BH-2 transmitted light microscope using a magnification of 500x. Using these methods, microfossils were then identified, classified, and examined for signs of possible use-wear.

RESULTS

Of the 110 samples analyzed, 63 (57%) yielded some type of microremain (Table 1). Numerous starch granules and fungal spores were recovered from the dental calculus samples. Some phytoliths and pollen grains were recovered, but they were few in number. In addition, microremains of water organisms: diatoms, chrysophytes, and sponge spicules, were also recovered, but rare.

Phytoliths

Phytoliths are microscopic silica structures found in certain parts of plants, including stems, leaves, and roots (Pearsall 2010:356). Phytoliths were present in 32 (29%) of the samples (Figure 1), and morphological characteristics suggest the presence of cool and warm season grasses, sagebrush, and squash. Interestingly, no maize phytoliths were identified and the lack of these could suggest the use of nixtamalization, a process in which corn is soaked and cooked in an alkaline solution (i.e. limewater), destroyed any of these microfossil silicates (Yost 2015). Maize subjected to the nixtamalization process has several benefits over unprocessed grain: it is more easily ground; its nutritional value is increased; flavor and aroma are improved; and mycotoxins are reduced.

Starch Granules

Starch granules were present in 47 (43%) of the samples (Figure 2). Starch was by far the most common microfossil identified, and maize starches represented the majority of this category (n=23). Grass grains (e.g., little barley) and one tuber starch were also identified. Lenticular granules showed signs of grinding and dry roasting (parching), while Zea mays granules showed signs of grinding, dry roasting, and fermentation (Figure 3). Large numbers of grinding tools have been found at Paquime and at surrounding sites (Searcy and Pitezel 2014; Whalen and Minnis 2015:54), and it is likely that many of the starch grains with evidence of damage from grinding, especially maize, were ground on manos and metates.

Microfossils Present in Dental Calculus

Microfossil Type	Total	Microfossil Type	Total	Microfossil Type	Total
STARCHES		**PHYTOLITHS**		**POLLENS**	
lenticular: Hordeum/Elymus (Pooideae)	16	trapeziform sinuate : C3 grass	1	unknown	3
spherical: grass seed (including maize)	5	keeled rondel: C3 grass	3	grass (Poaceae)	1
polyhedral: Maize (Zea mays)	32	angular keel: cf. Phalaris sp.	1	Pinus sp.	5
cluster: Maize (Zea mays)	2	Stipa-type bilobate: C3 grass	1	unknown: check local Prosopis sp.	2
irregular - possibly cooked: cf. Maize	13	rondel-pyramidal: C3 or C4 grass	3	**FUNGUS**	
swollen (partially gelatinized)	17	rondel-horned: typically C4 grass	2	Ustilago maydis fungal spore (corn smut)	4
cluster, swollen (partially gelatinized)	1	bilobate: C4 (Panicoideae) grass	4	Ustilago sp. fungal spore (corn smut)	3
cluster, possibly cooked	2	cross: C4 (Panicoideae) grass	1	Valsaria sp. ascospore (Fabaceae bark fungus)	1
fermentation/grinding damage	284	saddle: C4 (Chloridoideae) grass	18	Fungal spore	29
fermentation/grinding damage, fragment	75	elongate: grass/sedge epidermis	14	Fungal spore cluster	11
cluster, fermentation/grinding damage	44	bulliform: grass leaf	9	Plant/algae/fungal hairs or hyphae	1
triang. frags. (cf. gelatinized & fermentation)	57	trichome: grass or sedge	8	**DIATOMS/ALGAE**	
w/eccentric hilum: plant root	1	elongate facete: cf. Cucurbitaceae	2	Diatom aggregate (cf. marine) and starch granules	2
compound (double)	24	polyhedral faceted: woody plants	4	Diatom aggregate (Aulacoseira sp.) and starch	1
elongted with fissure: unknown	1	**OTHER**		Diatom: Aulacoseira sp. - mostly lakes	20
FIBER		Insect chitin	2	Diatom: pennate form - soils, ponds, lakes, rivers	1
Fiber (modern or ancient?)	55	Animal hair	1	Chrysophyte cyst: lake algae	1
Woody plant tissue w/bordered pits	4	Yellow flakes (unknown origin)	-	Sponge spicule	1

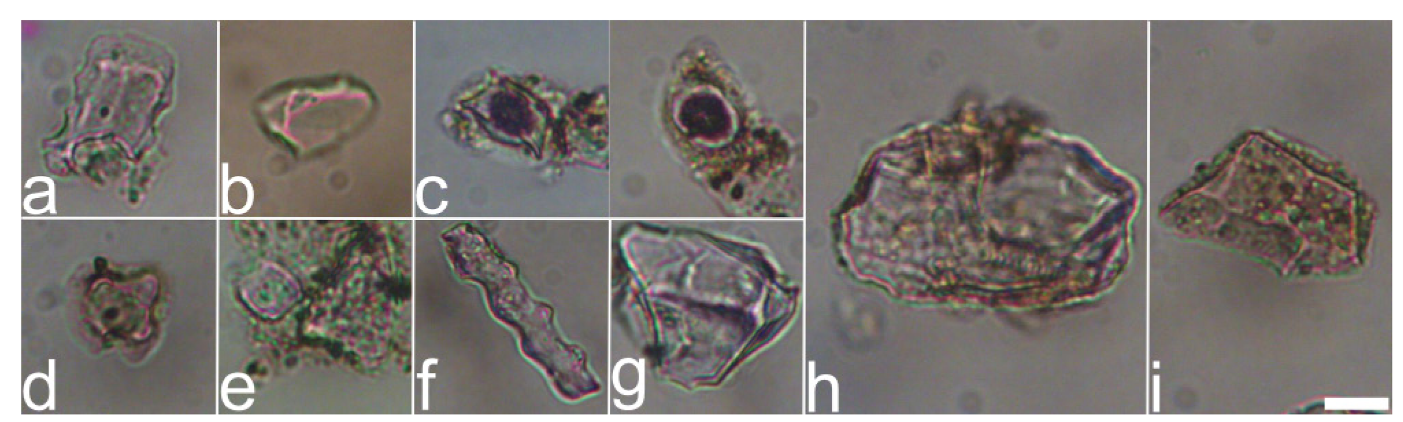

Figure 1. Micrographs of several phytoliths recovered from dental calculus samples. White scale bar equals 10 µm. a) Broken trapeziform sinuate diagnostic of C3 Pooideae grass leaves. b) Keeled rondel diagnostic of C3 grasses, and possibly derived from canarygrass (Phalaris sp.). c) Pyramidal rondel in side view (left) and plan view (right), typical of many grasses (Poaceae) and could be derive from maize (Zea mays). d) Cross phytolith typical of C4 Panicoideae grasses such as maize and others. e) Saddle phytolith diagnostic of C4 Chloridoideae grasses. f) Elongate echinate phytolith derived from either grass or sedge (Cyperaceae) epidermis. g) Polyhedral facetate phytolith derived from woody plant tissue (Sample 62). h-i) Hemispherical facetate phytoliths possibly derived from squash family (Cucurbitaceae) exocarp tissue (Samples 101 and 104, respectively). i) Facetate phytolith likely derived from squash (Cucurbita sp.) (Image and caption text from Yost 2015).

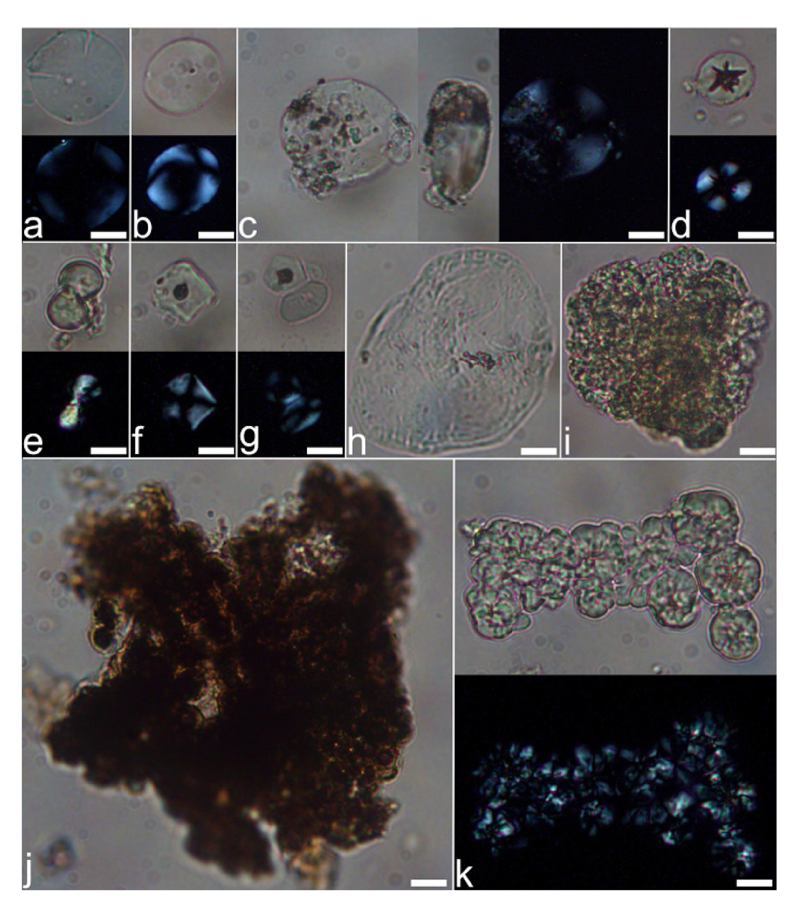

Figure 2. Selected starch granules recovered from dental calculus. White scale bars equal 10 µm. Dark field images are the granules viewed using cross-polarized light. a) Lenticular (in cross-section) starch granule diagnostic of Hordeum or Elymus grains. Cracks around the margin may be damage from grinding. b) Starch granule with eccentric hilum, typical of root/tuber starch. c) Lenticular starch granule most likely derived from Hordeum and exhibiting damage typical of dry roasting (parching). The middle image is a rotated view of the granule. d) Spherical starch granule typical of grass seed, including maize (Zea mays), and exhibiting possible cooking damage. e) Compound starch possibly from grass seed. f-g) Polyhedral starch diagnostic of maize. h) Gelatinized starch likely from cooking (sample 5). i) Possible cluster of gelatinized starch granules (sample 13). j) Cluster of hundreds of polyhedral (maize) starch granules (sample 2). k) Cluster of gelatinized starch, most likely from maize (sample 79).(Image and caption text from Yost 2015).

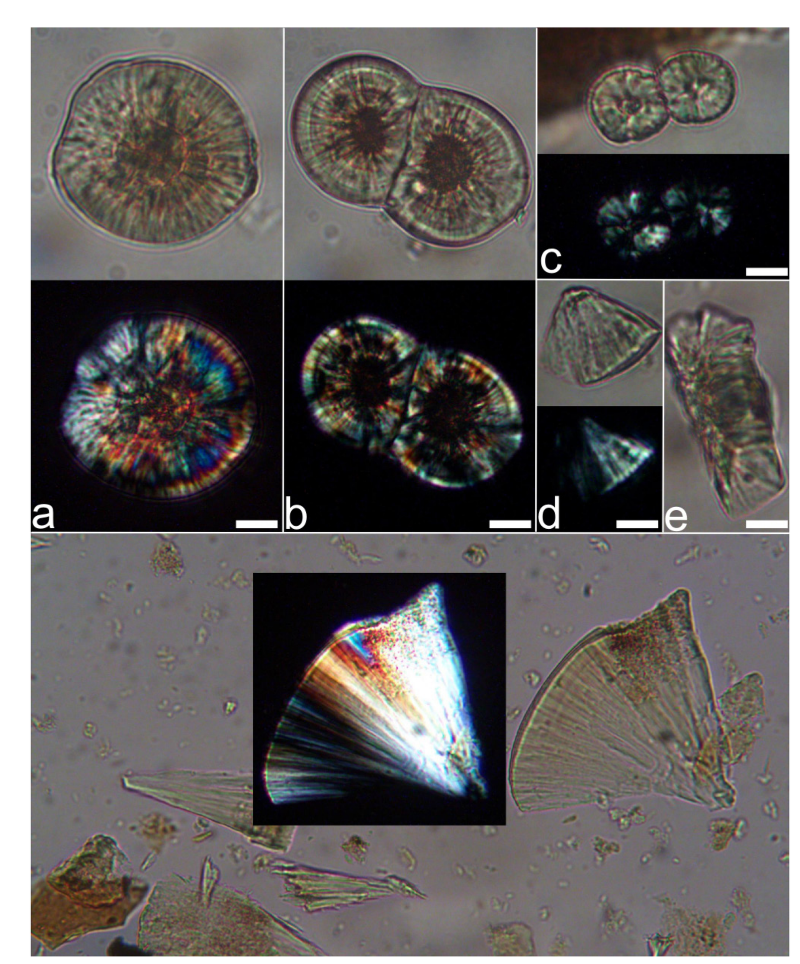

Figure 3. Micrographs of selected starch granules with grinding and fermentation damage. Dark field images are the granules viewed using cross-polarized light. White scale bars equal 10 µm. a-c) Damaged grains that are enlarged, but remain spherical, indicating fermentation under limited heating (a and b sample 79, c sample 62). d-f) Damaged grains that are slightly to greatly enlarged, flattened and often fragmented (sample 95), suggesting swelling and partial gelatinization before striae formation of the amylose, and suggesting significant heating during fermentation. e) The large fragment from (f) rotated to show the flattened morphology typical of this type of starch damage.(Image and caption text from Yost 2015).

Diatoms

Microremains of water organisms: diatoms, chrysophytes, and sponge spicules, were also recovered, but rare. *Aulacoseira sp.* diatoms identified in some of the samples are found in freshwater bodies of water, including ponds and lakes, and can be incorporated into food during processing or directly from drinking water. Their presence may suggest a riverine or lacustrine aspect to ancient diets, or may simply be evidence of water consumption from nearby rivers and springs. Additionaly, a large mass of diatoms, starch, and other org matter was identified. These diatoms could be of marine (ocean) or brackish water type.

Fungal Spores

Corn smut fungal spores (Ustilago maydis) were present in six (6%) of the samples, and other types of fungal spores were present in 13 (12%) of the samples. Corn smut was known to be eaten by people in the Americas, past and present, and the large galls that form on maize kernels are quite nutritious (Yost 2015). Huitlacoche is the Aztec term for this food and has a flavor similar to mushrooms.

Other

Pollen grains were present in 10 (9%) of the samples, including those from pine trees, which grow in the mountain forests of the Sierra Madre to the west. Ingestion of these grains was likely inadvertent due to the ubiquitous nature of pine pollen in this environ. Thin yellow flakes of unknown origin were also present in 44 (40%) of the samples, but Yost (2015) could not determine the organic matter from which they derived. Woody plant vascular tissue fragments in four samples also suggest possible medicinal uses of bark or the incorporation of wood fibers into food or nixtamal.

CONCLUSIONS

Based on our results, maize seems to have been the most common plant food represented in our samples from Casas Grandes. Cool season C3 grasses were also found and many lenticular starch grains showed signs of grinding, dry roasting, and cooking. Corn smut (Ustilago maydis) was found as well, which is unsurprising as this fungal pathogen has been determined to have originated and evolved along with maize domestication. There is strong evidence, ethnographic and archaeological, for the intentional consumption of corn smut (Valverde, et al. 1995), although unintentional calculus contamination is still a possibility.

Given the high amounts of maize and grass starch granules found, the near complete absence of phytoliths is interesting. As mentioned, the absence may suggest the use of nixtamalization to process the grains, which would eliminate the phytoliths from the microfossil record.

Perhaps the most interesting results were evidence for maize fermentation. Corn beer, or *chicha* has been recorded elsewhere in South and Central America. Three samples all showed damage resultant of the fermentation process. The granules exhibited striae from grinding, were swollen, or were gelatinous. Moreover, the three samples exhibit damage from three distinct heating temperatures, perhaps representing the entire chicha-making process. Whalen and Minnis (2015:51) have found some evidence of possible fermentation at a site west of modern-day Mata Ortiz. They found a number of pottery fragments from large vessels with "interior pitting that might have been the result of erosion due to the fermentation of a beer or from the alkaline treatment of large amounts of corn."

Yost (2015) suggests that "the presence of two marine diatoms in two diatom-starch aggregates indicates that fish and/or other marine aquatic resources were utilized." While the presence of diatoms is not surprising, it may suggest other food sources not commonly accounted for in the literature or the archaeological record.

Even more exciting are the complementary data being generated by other scholars concerning Casas Grandes health and diet. Katzenberg et al. 2015 have conducted preliminary analyses of skeletal morphometrics, isotopic characterizations, DNA, and paleopathologies of several of the same individuals studied in this research. We expect that these data will enhance our findings and provide unique dietary, genetic, health, and nutritional information regarding those who inhabited this desert region of Chihuahua.

REFERENCES

Boyadjian, Célia Helena C., Sabine Eggers, and Karl Reihard
2007 Dental Wash: A Problematic Method for Extracting Microfossils from Teeth. *Journal of Archaeological Science* 34:1622-1628.

Katzenberg, M. Anne, Kyle Waller, Adrianne Offenbecker, Courtney McConnan Borstad, and Ana Morales
2015 Jane's Influence: Violence, Mobility, and Social Organization at Casas Grandes. Paper presented at the 48th Annual Chacmool Archaeology Conference, Calgary.

Pearsall, Deborah
2010 *Paleoethnobotany: A Handbook of Procedures*, Second Edition. Left Coast Press, Walnut Creek, California.

Searcy. Michael and Todd Pitezel
2014 Exploring the First Ground Stone Quarry Discovered in the Casas Grandes Region Using Ethnoarchaeology. Paper presented at the 79th Annual Meeting of the Society for American Archaeology, Austin, Texas.

Valverde, M. E., O. Paredes-Lopez, J. K. Pataky and F. Guevara-Lara
1995 Huitlacoche (*Ustilago maydis*) as a food source--biology, composition, and production. *Critical Reviews in Food Science and Nutrition* 35(3):191-229.

Minnis, Paul E., and Michael E. Whalen
2015 Ecology and Food Economy. In *Ancient Paquime and the Casas Grandes World*, edited by Paul Minnis and Michael Whalen, pp. 41-57. University of Arizona Press, Tucson.

Yost, Chad
2015 *Microfossil analysis of dental calculus from Casas Grandes, Chihuahua, Mexico*. PAST Technical Report No. 15014.