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New insights into the consumption of maize and other food plants in the pre-Columbian Caribbean from starch grains trapped in human dental calculus

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Abstract

In a first region wide study, starch grains from human dental calculus from the pre-Columbian insular Caribbean (dating to ca. 350 B.C. – A.D. 1600) are used to identify important plant foods in the diet and to assess potential dietary differences related to age or sex. Results give important insights into pre-Columbian maize (*Zea mays*) consumption throughout the region, confirming recent studies that indicate that maize was more commonly consumed in the insular Caribbean than originally thought. No age or sex based differences in maize consumption were found. Furthermore, based on the results of new experiments regarding grinding and pressure damage to starch grains, it is clear that maize in the Caribbean was ground, baked and consumed as bread as was the case in large parts of the mainland. Based on our results we tentatively suggest maize consumption in the Caribbean was at least in some cases associated with feasting and ceremonial activities. The variety in other plant foods identified (mostly tuberous root crops) shows that the pre-Columbian inhabitants of the region consumed a broad spectrum, but locally variable diet in which a variety of root crops functioned as staple crops, including marunguey (*Zamia* sp.) and sweet potato (*Ipomoea batatas*). We found no indications for the traditionally assumed heavy reliance on manioc (*Manihot esculenta*) cultivation in the region.

Keywords: Starch grains; Dental calculus; Diet; Plant microremains; Caribbean; pre-Columbian; maize

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1. Introduction

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Our understanding of pre-Columbian plant exploitation in the Caribbean is somewhat convoluted by the region's (sub)tropical climate, which hampers the preservation of botanical remains. Despite this, paleoethnobotanical research over the past few decades has immensely improved our understanding of pre-Columbian plant use in the region, bringing to light a long history of Amerindian plant exploitation and cultivation, characterized by intensification and diversification of plant reliance over time, from the initial settlement of the region around 5500 B.C. to the first contact with Europeans in A.D. 1492. Based on faunal and botanical remains, ethnohistoric accounts, ethnographic analogies, and archaeological artifacts associated with root crop horticulture, it is widely understood that the first human management of plants in the Caribbean started in Preceramic times (~5500 – 400 B.C.), and intensified during the Early Ceramic Age (400 B.C. – A.D. 600/800). By the Late Ceramic Age (A.D. 600/800 – 1492) pre-Columbian Caribbean Amerindians subsisted predominantly on home gardening, cultivating staple food plants originating in the South American mainland such as manioc (*Manihot esculenta*) and sweet potato (*Ipomoea batatas*). These were supplemented by a range of other fruits and vegetables, next to meat and fish (Newsom and Pearsall, 2003; Newsom and Wing, 2004; Rouse, 1992; Sturtevant, 1969).

Recent microbotanical studies of residues on stone tools and pottery in the region have brought to light some interesting and unexpected aspects of pre-Columbian plant consumption, including evidence suggesting that maize (*Zea mays*) – hitherto thought to have been a relatively late introduction to the region and considered to have been of minor importance to the overall diet – was used early in the history of occupation of the islands and may have comprised an important component of the broad-based mixed horticulture practiced during the entire pre-Columbian period (Berman and Pearsall, 2000, 2008; Pagán-Jiménez, 2007, 2009, 2011; Pagán-Jiménez et al., 2005; Pagán-Jiménez and Rodríguez-Ramos, 2007; Pearsall, 2002). Furthermore, cooking and preparation implements known as *burenes* or clay griddles traditionally associated with manioc horticulture have yielded starch grains belonging to a variety of plants (including maize), and even fatty residues of meat and fish (Rodríguez-Suárez and Pagán-Jiménez, 2008). Surprisingly, no manioc microremains were recovered from these implements and their minimal presence in ceramic, coral and stone tools from sites spanning the pre-Columbian occupation of the region has led Pagán-Jiménez to question the degree of importance which has conventionally been attributed to this crop in Caribbean archaeology (Berman and Pearsall, 2008; Pagán-Jiménez, 2009, 2012).

These studies are changing our view of pre-Columbian plant use in the Caribbean, most significantly in showing that maize, a crop of considerable importance in

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contemporaneous mainland societies, may have been more common than previously thought. Macrobotanical evidence for maize consumption in the region is scarce, and excepting recent ancient starch grain studies, it has a strikingly low signal in the archaeological record, raising the question of this crop's importance in pre-Columbian Caribbean subsistence (Lane et al., 2008). The precise social and cultural contexts of maize consumption are still sketchy. One way of helping resolve this issue is by studying individual plant food consumption patterns.

<<Figure 1>>

Map of the Caribbean showing the sites used in this study: 1. El Chorro de Maíta; 2. Juan Dolio; 3. El Cabo; 4. Punta Macao; 5. Maisabel; 6. Tutu; 7. Kelbey's Ridge; 8. Anse à la Gourde; 9. Point de Caille; 10. Escape; 11. Manzanilla; 12. Malmok; 13. Tanki Flip; 14. Canashito

Here, we present the results of the first regional starch grain analysis of 30 samples of dental calculus adhering to human teeth from 14 pre-Columbian sites in the insular Caribbean, i.e., the islands in the Caribbean Sea as opposed to the mainland areas of the Caribbean which surround the islands (Figure 1, Table 1).[1] The results pertain to the period between ca. 350 B.C. – A.D. 1600, a time frame in which important social and cultural changes associated with multivectorial pan-Caribbean interactions took place. These developments include the foundation, expansion and consolidation of regional social networks, in which material goods were exchanged across large distances in order to uphold social and ancestral ties (Mol, 2011; Rodríguez-Ramos, 2010; Rodríguez-Ramos and Pagán-Jiménez, 2006). Important sociopolitical developments involving increased settlement stability, development of increased independency in community resource procurement, and growing regional settlement hierarchy in the Late Ceramic Age were grounded in these social interaction networks (e.g., Crock and Carder, 2011; Hofman and Hoogland, 2011; Samson, 2010). This regional approach is unique in that it reveals highly detailed individual plant food consumption patterns across space and time, allowing us to study age and sex differences and shed light on the overall degree of importance of various crops. Patterns of damage to the starch grains, indicating the use of heat and pressure, give important insights into processing and cooking techniques. Our findings on the processing of starches for consumption are based in part on the results of a grinding experiment (Appendix A) aimed at documenting patterns of grinding

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[1] The final project includes 60 samples of dental calculus from various sites throughout the region. Here, we present the results of the analysis of the first 30 samples.

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damage in maize starch grains at different developmental stages. The results of this experiment with modern maize starches shed new light on pressure damage to starches, contributing to our knowledge of archaeological starch grains in general.

<<Table 1>>

Samples by site, age, sex, radiocarbon date and cultural assignment. LCA= Late Ceramic Age. ECA= Early Ceramic Age. * No starches recovered. † Site dating based on relative chronology (ceramic typology).

1.1 Ancient starch grain analysis of dental calculus

Dental calculus is the mineralized form of dental plaque, a biofilm containing food remains and bacteria, which forms naturally on the teeth, and when it is not removed may mineralize into calculus (Hillson, 1996; Lieverse, 1999). During the formation of dental plaque, food particles such as plant microremains may become trapped, and are protected from chemical breakdown by salivary amylase (Juan-Tresserras et al., 1997). Once plaque mineralizes into calculus, these microremains are protected within the robust mineral matrix, aiding their preservation (Cummings and Magennis, 1997; Hardy et al., 2009). Starch grains recovered from human dental calculus provide an excellent opportunity to better understand plant use, crop cultivation, and culinary practices. They can be used to distinguish between food plants versus plants used for non-food products such as ointments and pigments, as in most cases plant microremains found in the mouth will have derived from the food. Also, unprocessed or raw plants which would not leave residues on tools, can be identified (Cummings and Magennis, 1997; Hardy et al., 2009; Henry and Piperno, 2008; Henry et al., 2011; Piperno and Dillehay, 2008).

Even so, starch grains trapped in dental calculus do not simply reflect the range of plant foods consumed by an individual, as consumption of a plant does not guarantee that its starches are preserved in the calculus. Likewise, the frequency of consumption of particular plants is hard to predict; presumably frequent consumption raises the chances of starches becoming trapped. However, there are differences in starch production in different taxa and different plant organs, making any prediction of frequency of consumption based on the number of starches recovered highly unreliable. In many cases, starches simply can not be identified, as not all plants produce diagnostic starches. In addition, the process of calculus formation is still poorly understood. Various factors, including diet, oral hygiene, salivary flow and genetics influence the rate of calculus deposition, and as a consequence the time

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frame of accumulation in individuals is hard to predict (Hillson, 1996; Lieverse, 1999). Generally, medium to large calculus deposits are thought to have accumulated over at least a number of years (Brothwell, 1981; Li et al., 2010; Henry and Piperno, 2008; Piperno and Dillehay, 2008).

1.2 Maize in the pre-Columbian Caribbean

Maize was originally thought to have been a late pre-Columbian introduction to the Caribbean archipelago, most likely from the South American mainland, with macrobotanical remains currently known only from Late Ceramic Age (Chican Ostionoid; A.D. 1000 – 1500) contexts in the Greater and northern Lesser Antilles (Pearsall, 2002; Newsom and Deagan, 1994; Newsom and Wing, 2004; Rouse, 1992; Sauer, 1966). It was purportedly not consumed as a staple food in the Caribbean, and some suggest it was consumed only in ritual or restricted high-status contexts (Newsom, 2006; Newsom and Deagan, 1994; Newsom and Pearsall, 2003; Newsom and Wing, 2004; see also Piperno, 2002). Its very low signal in the archaeological record has led Newsom (2006) to propose the plant was consumed only as a supplementary food in its immature state. Sixteenth-century chronicler Gonzalo Fernández de Oviedo's description of the use of maize in Hispaniola and the other Caribbean islands in its green state or as roasted kernels adds credence to this idea (Fernández de Oviedo, 1851 Vol. I: 266). Pearsall's (2002) research has shown that maize likely comprised an important component of the broad-based mixed horticulture practiced during the Late Ceramic Age occupation of the region. Furthermore, evidence of maize agriculture has recently been found in the form of pollen grains in lake sediments from the interior of the Dominican Republic, providing a secure dating of cal. A.D. 1060, and hinting at possible large scale cultivation of maize (Lane et al., 2008).

While the bulk of macrobotanical evidence for maize in the Caribbean pertains to the Late Ceramic Age, some argue that it may have been introduced to the region at a much earlier stage, perhaps even by the first migrants to the islands. Recent research is steadily uncovering more evidence of 'early maize consumption' in the region. Starch grains on chert microliths have shown that by at least A.D. 800 and perhaps earlier, the inhabitants of the Bahamas used maize, along with chili (*Capsicum* sp.) and perhaps manioc and other starchy plants such as marunguey (*Zamia* sp.). Significantly, the investigators conclude that maize was *"part of a broad-based diet […] and was not a supplemental or "curiosity" crop"* (Berman and Pearsall, 2008: 194). Analyses of stone grinding implements from sites in Puerto Rico, Vieques, the Dominican Republic, Cuba, and Saba have revealed that maize was being

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consistently exploited as early as 3000 B.C. up to the contact period around A.D. 1500 (Pagán-Jiménez, 2007, 2009, 2011, 2012; Pagán-Jiménez et al., 2005; Pagán-Jiménez and Rodríguez-Ramos, 2007).

Stable isotope analysis of carbon and nitrogen in human bone has provided evidence of differential dietary practices within and between three Ceramic Age Puerto Rican sites (Pestle, 2010). At all three sites a notable enrichment in $\delta^{13}C_{ap}$ values was observed, which Pestle interprets as showing that C_4/CAM plants comprised a moderate to substantial portion of the diet (Pestle, 2010; Smith and Epstein, 1971; see also Stokes, 1998). The consumption of C_4/CAM plants, by inference including maize, at all three sites was not restricted to a small (elite) group. Some of the samples dated to a few centuries earlier than previously accepted dates for the integration of maize into the Caribbean islands. However, while considering the possibility that maize was the cause of $\delta^{13}C_{ap}$ enrichment in over 200 human skeletons, Pestle (2010) takes care to note a number of other C_4/CAM plants, which could have been the cause, including amaranth (*Amaranthus* sp.), century plant (*Agave antillarum*), pineapple (*Ananas comosus*) and prickly pear cactus (*Opuntia* sp.).

2. Materials and methods

Thirty human teeth from burial contexts in 14 sites throughout the Caribbean archipelago were chosen (Figure 1, Table 1). Based on previous analyses of the dental wear and pathology in these assemblages, it is clear that these populations were consuming very large amounts of starchy foods, along with marine and terrestrial proteins (e.g., Mickleburgh, 2007, 2011). The samples derive from sites dating mostly to the Late Ceramic Age, with the exception of Malmok and Canashito, which date to the Preceramic or Archaic Age of the southern insular Caribbean, and Escape, which dates to the Early Ceramic Age. The site of El Chorro de Maíta dates to the Late Ceramic and Early Contact period. The multi-component sites of Maisabel and Tutu offered the opportunity of sampling Early Ceramic Age and Late Ceramic Age individuals from the same site. Although we strived to include equal numbers of early and late individuals overall, there is a general lack of early (especially Archaic) skeletal material from secure archaeological contexts.

In total, calculus from teeth of 11 males, 10 females, 4 juveniles, and 5 adults of unknown sex was analyzed. Where possible, an adult male, an adult female and a juvenile were selected for each site. In some cases this was not possible due to the lack of calculus or dental material from individuals of known age and sex. Also, where possible, individuals with known radiocarbon dates were selected.

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Extracting protocol for dental calculus consisted first of choosing the area on each tooth with the largest calculus deposit. Photographs were taken of the sampling area on each tooth, prior to and after extraction. Each tooth was washed separately using distilled water and new sterilized dental brushes, and calculus was extracted using a sterilized dental pick. The weight and volume of the individual samples was recorded. Subsequently, they were treated with a 10% solution of HCl at room temperature for 24 hours. Distilled water was added to each microcentrifuge tube containing the individual samples; these were agitated manually for 20 seconds and then centrifuged (4500 rpm for 3 min). This step was repeated twice. The samples were mounted on microscope slides, adding a half drop of glycerol to each, and examined with an Olympus BH-2 microscope with polarization capacity at 40X. Starch grains were counted, described and photographed, and compared to Pagán-Jiménez' reference collection (Pagán-Jiménez, 2007) comprising starch grains from 68 genera and 61 species of wild, domesticated, and cultivated species from the Antilles, continental tropical America, and parts of the Old World, in order to identify taxa. Published literature on diagnostic criteria for starch grains was also consulted (Pearsall et al., 2004; Piperno and Dillehay, 2008; Piperno and Holst, 1998; Reichert, 1913). Sample selection was performed by Mickleburgh at Leiden University, while starch grain extraction and analysis was performed by Pagán-Jiménez at the Universidad de Puerto Rico (Río Piedras).

Patterns of damage to the starch grains, indicative mainly of the manner of preparation of the plants for consumption were documented according to a reference collection of starch grains resulting from various processing experiments (Appendix A), and were also compared to previously documented patterns of damage (Babot, 2003; Dorsey et al., 2009; Henry et al., 2009; Lamb and Loy, 2005; Piperno et al., 2004). Thus, secure and tentative identifications of starches in this study are based on diagnostic and/or distinctive features described elsewhere and on the results of new experiments by Pagán-Jiménez (Appendix A). Specific morphometric features used are shape, size, presence and location of the hilum within the granule, presence and appearance of fissures, presence and type of pressure facets, presence and appearance of lamellae, and in some cases the appearance and projection of the Maltese cross. A detailed discussion of the diagnostic criteria used for the identification of some of the taxa in this study is presented in Appendix B.

3. Results

A very high success rate (90%) was obtained for the extraction and identification of starch grains from dental calculus (Table 2). In all but 3 samples, starch grains were recovered

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and identified. The number of starch grains per sample is slightly lower than is the case in similar studies in other regions (e.g., Hardy et al., 2009; Henry et al., 2011; Henry and Piperno, 2008; Li et al., 2010; Piperno and Dillehay, 2008). Adaptation of the extraction technique[2] did not improve the retrieval rate, and as the method we have used is comparable to those used in other studies (i.e., Hardy et al., 2009; Henry and Piperno, 2008), we feel that the lower retrieval rate is most likely the result of poor preservation of the material (most of the remains, including the calculus deposits, were very poorly preserved).

<<Table 2>>

Identified taxa per sample and the number of individual starch grains identified. Site abbreviations are as follows: AAG= Anse à la Gourde, CB= El Cabo, CM= El Chorro de Maíta, CS= Canashito, ES= Escape, JD= Juan Dolio, KR= Kebey's Ridge 2, MB= Maisabel, MK= Malmok, MZ= Manzanilla, PC= Point de Caille, PM= Punta Macao, TF= Tanki Flip, TT= Tutu. Other abbreviations are: cl= cluster of starches, P= pressure, F= fermentation, H= heat, W= water. Thus H/W= heat in the presence of water, and P/H?= pressure, possibly with heat. * Starch clusters are not included in the totals, as the precise number of starches in the cluster is unknown

A large number of tuberous root plants were identified (Appendix B), including marunguey (*Zamia* sp., *Zamia pumila*, cf. *Zamia erosa*), sweet potato, cocoyam (*Xanthosoma* sp., *Xanthosoma* cf. *sagittifolium*), arrowhead (cf. *Sagittaria* sp.) and manioc (Figure 2). The latter was represented by a single starch grain retrieved from a female individual from Malmok, Aruba (burial 10). The rhizomes Cannaceae (which includes the economic species such as achira or gruya), Marantaceae (for which arrowroot is one of the better-known economic species) and Calathea (*Calathea* sp.) were also tentatively identified. A number of legumes were identified, including *Canavalia* sp., common bean (*Phaseolus vulgaris*), and other wild legumes (e.g., Fabaceae, Leguminoseae). Finally, in almost half of the samples which yielded starch grains (13 individuals from 11 sites), maize starch was discovered.

<<Figure 2>>

Microbotanical residues recovered from human dental calculus. Rows A-F, starch grains and a raphide. Row G, phytoliths. Rows A and B: *Zea mays* starches. (A1, A2, A3, B1, B2, B3[2])

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[2] Only distilled water was used to dissolve the samples instead of HCl, in order to establish whether starches were being lost during sample processing.

securely identified *Zea mays*, (A4, A5, B3[1]) tentatively identified *Zea mays*. Row C: (C1, C4, C5) *Zamia* sp. (secure); (C2) *Zamia pumila* (secure); (C3) *Zamia erosa* (tentative). Row D: (D1) *Phaseolus* sp. (secure); (D3, D4) *Ipomoea batatas* (tentative); (D5) *Manihot esculenta* (secure). Row E: (E1) *Calathea* sp. (secure); (E3) *Sagittaria* sp. (tentative); (E4) *Xanthosoma* sp. (tentative); (E5) Cannaceae (tentative). Row F: (F1) *Xanthosoma* sp. starch cluster (secure); (F2) transitory-like unidentified starches (probably from leaves); (F3, F4) pressure damage; (F5) unidentified raphide. Row G: (G1, G2, G3) globular echinate phytoliths likely from palm stems, leaves or fruits. CD1, large central depression; CD2, small central depression; R, bright ring; Db, double-border; F, general fissures; F1, radial fissures or striations; F2, radial to asymmetric fissures; P, pressure facets; H, hilum; L, lamellae; Rs, rough topography; Fs, fold/shadow (likely produced by boiling). All scale bars are 30µm. Dark field micrographs with cross polarized light are showing general characteristics of Maltese crosses and correspond to the same starches to its immediate left.
(C1 and G3) Chorro de Maíta; (C2) El Cabo; (A2) Juan Dolio; (G5) Punta Macao; (A1, E1, E2, E4, G4) Anse á la Gourde; (G1, G2) Escape; (D3) Kelbey's Ridge; (A3, C3, E3) Maisabel; (D5, F1) Malmok; (F4) Canashito; (A4, A5, B1, C4, C5, F5) Manzanilla; (B3) Point de Caille; (B4, B5, D4, E5, F2, F3) Tutu.

Many of the starch grains displayed alterations in morphometric characteristics and patterns of damage consistent with pressure and heat treatment (Dorsey et al., 2009; Henry et al., 2009; Figure 2, Table 2, Appendix A). Several starch grains (Table 3) displayed substantial enlargement when compared to previously published data (e.g., Pagán-Jiménez, 2007). Most enlarged starch grains belonged to maize, and their sizes correspond to the size ranges registered in the grinding experiment, where modern maize starch grains were submitted to intensive grinding (Table 2 in Appendix A).

<<Table 3>>
Size ranges of starch grains identified in this study.
* Starch clusters are excluded

In most cases it was possible to distinguish between damage patterns consistent with heat treatment in the presence of liquid (i.e., boiling; as evidenced by the presence of folds and large central depressions) versus heat treatment without large amounts of liquid (i.e., baking, toasting, parching, popping, etc.; as evidenced by a 'crushed' appearance). Only three

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cases damage consistent with boiling were identified, while dry heat treatment was fairly common (n=39).

Patterns of damage consistent with pressure treatment were observed in all but two of the individuals who yielded starch grains (Figure 2, Table 2), with 70.2% (99/141) of all starches showing signs of pressure treatment. Based on the results of the grinding experiment, the damage was mostly found to be related to grinding or pounding (Appendix A). Radial as well as asymmetric striations and fissures in the grains were the most common signs of damage observed in the maize starches and other recovered starches (see also Figure 2 in Appendix A). Other patterns consistent with pressure damage were the presence of a bright ring around the hilum area (mainly in maize starches, but also in cocoyam and common bean), the enlargement of some starch grains, and the formation of a central depression in the hilum area as registered in the grinding experiment. Together with other types of central depressions associated with heat (i.e., “folds” or “shadows” *sensu* Henry et al., 2009) and grinding damage patterns (Appendix A), this suggests that in the case of maize the kernels were intensively ground or pounded in a mature or dry state (previously soaked) and subsequently baked, indicating that the starch was most likely consumed as bread, as opposed to in its immature or green state.

As discussed above, recent research has steadily been pushing back the date at which there is paleoethnobotanical evidence for maize consumption in the pre-Columbian Caribbean. Our results show that maize was consumed by two individuals (burials 16 and 30; see Tables 1 and 2) dated respectively to the Early and Late Ceramic Age phases of occupation at Tutu, St. Thomas. Previous research based on macrobotanical remains at the site indicated maize consumption in the Late Ceramic Age (cal. A.D. 1150 – 1500) phase of occupation (Pearsall, 2002; Sandford et al., 2002).

The single individual from the Archaic period site of Canashito on Aruba yielded a relatively high number of maize starch grains, with evidence of grinding and baking. Although this individual has not yet been radiocarbon dated, a closely associated burial, was dated to cal. 350 B.C. – A.D. 150, and artifacts collected at the site and from the burial pits clearly put Canashito in the Preceramic or Archaic Age of the southern insular Caribbean (Dijkhoff and Linville, 2004; Versteeg et al., 1990; Wagenaar Hummelinck, 1959).

The identification of other domestic and wild taxa such as sweet potato, beans, cocoyam, marunguey, and wild legumes is consistent with previous starch grain research on lithic, ceramic and shell tools in Puerto Rico, Cuba, the Bahamas and the Dominican Republic (Berman and Pearsall, 2008; Pagán-Jiménez, 2011, 2012) where a broad spectrum, but locally variable diet has been proposed for different periods in the pre-Columbian occupation of the region.

Apart from starch grains a number of other inclusions were found in the calculus samples. Seven of the samples yielded spiny-globular echinate phytoliths which were tentatively identified as belonging to palms (Figure 2). Charcoal particles were found in 16 of the 30 samples. The presence of these particles deep in the mineral matrix of the calculus indicates that they are highly unlikely to be the result of post mortem contamination as previously suggested (Wesolowski et al., 2010). Instead, it appears that charcoal was present in the food or at least in the oral cavity of these individuals during life, perhaps as the result of cooking techniques. Baking or roasting foods directly in the fire or on glowing embers would result in the adherence of charcoal particles in and on the food.

4. Discussion

The large number of plants identified in the entire sample set, and at the individual sites, shows that these individuals consumed a large variety of cultivars, confirming earlier studies that have highlighted the variety in starchy plant foods being consumed in the region (Berman and Pearsall, 2008; Newsom and Pearsall, 2003; Newsom and Wing, 2004; Pagán-Jiménez, 2007, 2011). The variety of tubers and rhizomes identified in the sample is especially interesting when contrasted with the relative paucity of manioc, which was identified by only a single starch grain. Although considered a staple crop during the Ceramic Age in the insular Caribbean and northeastern South America, manioc has to date been extremely scarce in paleoethnobotanical remains studied in the region. Ancient manioc starches have been identified in a total of seventeen (10.2%) of 168 tools sampled so far from the insular Caribbean and French Guiana (Pagán-Jiménez, 2011). Of the 28 *burenes*, artifacts traditionally associated with the preparation and baking of manioc bread, only one revealed manioc starches. Interestingly, the stone ‘teeth’ of grater boards, also traditionally associated with manioc processing in the Caribbean and Venezuela, have yielded a variety of tuber and seed starches, but very little evidence of manioc (Berman and Pearsall, 2008; Pagán-Jiménez, 2009; Perry, 2005). The reasons for this scarcity of recovered manioc starches in the pre-Columbian neotropics are not well understood. As manioc starches have been successfully recovered and identified in a small number of cases, it is hard to assume that taphonomic or laboratory biases are the (sole) cause. Recent studies consistently suggest that manioc was

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simply a tuber crop of minor importance prior to European contact (Berman and Pearsall, 2008; Pagán-Jiménez, 2012; Perry, 2005; see also Appendix B).

The lack of manioc starches in our samples underscores previous suggestions that various other plants such as marunguey and sweet potato may have fulfilled the role of staple food traditionally attributed to manioc (Berman and Pearsall, 2008; Pagán-Jiménez, 2009, 2011). Our data support the view that a variety of root crops fulfilled that purpose in the subsistence economy.

The identification of marunguey in this study is illuminating. Previous starch grain analyses at various archaeological sites in the Greater Antilles revealed that the tuberous stem of this wild plant was an important food item throughout human occupation of the area (Pagán-Jiménez, 2011, 2012). So, the presence of marunguey starch grains at sites in the Greater Antilles is unsurprising as wild populations of this genus are known to have grown in limestone areas in Puerto Rico, Hispaniola, Cuba, Jamaica, the Cayman Islands, and the Bahamas for many millennia. Its presence in islands in the southern Caribbean, such as Aruba and Trinidad is highly interesting, however, as no wild marunguey populations currently grow there, and there is no botanical or palynological evidence for its presence prior to European contact. These islands share some important geological characteristics (limestone formations) with areas in the Greater Antilles, which are favorable for the development of wild populations of marunguey. Three different scenarios explaining these findings are posited: (a) the inter-island exchange of marunguey and introduction of this species into home gardens or agricultural plots beyond its natural range, (b) the inter-island exchange of marunguey (finished) food items, or (c) the presence of wild populations of marunguey in these southern islands during the pre-Columbian period. Needless to say this matter warrants further investigation.[3]

Extremely significant is the high frequency of maize starch. Almost half of the individuals in this sample consumed maize, often in its mature or dry state, ground and probably baked as bread. Combined with the presence of charcoal remains in many of the samples, this may be direct evidence of the *barbacoa* or barbecue cooking technique - where foods (including fauna) are baked in or over an open fire – described in ethnohistoric sources (Fernández de Oviedo, 1851 Vol. I: 559 and 561). The large number of individuals who yielded maize starch appears to confirm earlier suggestions that maize may not have been as restricted a food source as previously thought (Pagán-Jiménez and Oliver, 2008; Pagán-

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[3] For a more in depth discussion of the consumption of marunguey in the pre-Columbian Caribbean, see Appendix B.

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Jiménez, 2011; Pestle, 2010). Interestingly, none of the juveniles in the sample yielded maize starch, perhaps indicating at least some form of restricted access to the plant. However, based on such a small number of juvenile individuals (three who yielded starch grains) this is hard to support at this time. Excepting this, there is no evidence in the form of mortuary practices and/or grave goods, age, sex, or otherwise indicating that the individuals who yielded maize starch in this sample belonged to an elite class or had a different social status than those who did not yield maize starch. No significant differences were found between males and females. Besides, as discussed above, those lacking maize starch grains in their calculus did not necessarily avoid eating maize, as consumption of a plant does not guarantee that starch grains will be preserved in the calculus. This means that the actual number of individuals consuming maize may have been higher. However, as discussed above, the presence (or absence) of starch grains in dental calculus gives no reliable indication of the frequency of consumption. We are cautious to suggest that these individuals were consuming substantial amounts of maize, as there is some evidence which indicates maize consumption was minimal.

The spatial and chronological distribution of the samples containing maize in this study represent more evidence that access to the plant was not highly restricted, as our data show maize was consumed at 11 of the 14 sites represented, as early as 350 B.C. – A.D. 150 and as late as A.D. 1250 – 1600 (see Tables 1 and 2). It is clear that maize consumption in the insular Caribbean was a far more ubiquitous practice than once thought (*sensu* Newsom, 2006). However, at the moment it seems unlikely that the importance of maize in the subsistence economy was on a par with that of others crops, such as some of the root crops mentioned above. While our data seem to indicate maize consumption was not highly exclusive, some previous paleoethnobotanical and bone isotopic studies appear to show that it was most likely not consumed in great amounts as a staple crop.

At the site of Tutu, St. Thomas, the presence of macrobotanical remains, which included a tiny amount of charred maize kernels dated to the Late Ceramic Age phase of occupation, led Pearsall (2002) to suggest small amounts of maize were consumed, although it did not fulfill an important role in subsistence practices at the site. A study of phytoliths in soil samples found no evidence of maize production or consumption (Piperno, 2002), and the presence of Marantaceae, Palmae and squash (*Cucurbita* sp.) led Piperno to conclude that the vegetal portion of the diet consisted mainly of tubers and tree crops. A human bone isotopic study revealed a mean bone collagen δ^{13}C value of –15.50 ±1.80‰ (s.d. 2), and a mean bone collagen δ^{15}N value of 12.10 ±1.70‰, predominantly reflecting a large marine component in

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the protein portion of the diet and the consumption of reef and pelagic fishes (Norr, 2002). The mean human bone apatite carbonate $\delta^{13}C$ value is –10.30 ±2.40‰ (adjusted by Norr to 9.50‰), which Norr (2002) interpreted to reflect a diet intermediate between the C_3 plants in the food chain and marine, but which given our evidence for maize consumption at the site, may also reflect a very small component of C_4 plants in the diet. Comparison of our results with these previous studies thus indicates that C_4 plants (such as maize) may have been consumed in small amounts by the inhabitants of Tutu.

At the Late Ceramic Age site of En Bas Saline, Haiti, researchers discovered that maize macroremains were clearly associated with the centrally positioned high-status or elite area of the site, specifically the center where the cacique's (chief's) residence was situated. The remains were recovered from what appear to have been feasting pits or communal hearths (Deagan, 2004; Newsom, 1998; Newsom and Deagan, 1994), and the finds were interpreted as evidence for a distinct social significance of and potentially restricted access to maize. A similar setting has been suggested for Formative societies in Mesoamerica, where maize was considered a highly important and ritually significant plant, and was primarily consumed during communal feasts (Seinfeld, 2011).

As discussed above, recent stable isotope analysis at three Ceramic Age Puerto Rican sites has revealed that C_4/CAM plants likely comprised a large portion of the vegetal diet, with an estimated average of 47±8.1% of dietary energy being provided by C_4/CAM carbohydrates (Pestle, 2010). The results of this study indicated that maize was unlikely to have been a restricted or 'elite' food, but despite the considerable $\delta^{13}C_{ap}$ enrichment in the majority of individuals, maize was not thought to be a staple crop at any or all of these sites, as a number of other C_4/CAM plants likely contributed to the diet.

Considering the above discussion in the light of our own evidence, we suggest that maize consumption in the pre-Columbian Caribbean may have been associated mainly with communal activities such as feasting, as opposed to the more mundane and common consumption of staple foods in the non-public domain. The lack of evidence for highly restricted access to maize, combined with indications that only small to moderate amounts were consumed, supports the view that maize consumption may have been associated with the public domain, or in other words, communal ritual and ceremonial activities. A high-status or 'special' significance attached to the plant would have made it particularly suitable as a feasting food (Hayden, 1996), consumed only on special occasions, but by most (if not all) individuals. This scenario may go some way in explaining the paucity of macrobotanical maize remains in archaeological deposits in the region, as the currently identified remains of

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individual feasting events are scarce (Curet and Pestle, 2010; Crock and Carder, 2011; Deagan, 2004; Newsom and Deagan, 1994; Oliver and Narganes Storde, 2003). While numerous studies in the Caribbean have linked elaborately shaped and decorated pottery with communal feasting activities (e.g., Boomert 1999; Hofman, 1993), and recent research has focused on recognizing feasting activities from the composition and size of a site's faunal assemblage (Crock and Carder, 2011; but see also Curet and Pestle, 2010), defining the 'archaeological footprint' of individual feasting events in the region remains a challenge.

Finally, it is important to keep in mind that the status of maize, whether mundane or socially and ritually significant, may have been highly variable across the region, warranting more detailed investigation on a local scale.

5. Conclusions

Our results support previous findings that a broad spectrum, but locally variable diet was consumed throughout the Caribbean islands, from ca. 350 B.C. – A.D. 1600. In this broad spectrum a variety of root crops functioned as staple crops in the subsistence economy, including marunguey and sweet potato. We found no indications for the traditionally assumed heavy reliance on manioc cultivation in the region.

Our results also clearly show that maize consumption in the pre-Columbian Caribbean archipelago was not a highly restricted practice, and, consequently, does not appear to be associated with an elite or high-status class of people. Neither are there any indications of sex based differences in access to the plant. Maize was consumed by individuals dating to the Preceramic/Archaic Age, Early Ceramic Age, and Late Ceramic Age occupation of the region, mostly ground and baked as bread, instead of in its immature or 'green' state. Naturally, this again illuminates the issue of the general lack of macrobotanical evidence for maize consumption in the region. We suggest this may, among other things, be the result of an association of maize with communal feasting activities, which are currently rarely identified in the archaeological record of the region.

Finally, this study has highlighted the value of an approach geared toward exposing past human activity on an individual level. Future work, among other things, will focus on potential intra-site age and sex differentiation in plant food consumption.

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Table 1

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Site	Island/Country	Burial	Sex	Age	Radiocarbon date	Cultural assignment	Site dating	Source
Anse à la Gourde	Guadeloupe	196	Male	26-35		LCA	cal. A.D. 450-1350	Hofman et al., 2003; Weston pers. comm. 2010
Anse à la Gourde	Guadeloupe	197	Male	18-25		LCA	cal. A.D. 450-1350	Hofman et al., 2003; Weston pers. comm. 2010
Anse à la Gourde	Guadeloupe	2217	Female	18-25	cal. A.D. 1310-1440	LCA	cal. A.D. 450-1350	C.L. Hofman and M.L.P. Hoogland, pers. comm. 2011; Weston pers. comm. 2010
El Cabo	Dominican Republic	85-40-17	Unknown	adult		LCA	A.D. 600-1504[†]	Samson, 2010
El Chorro de Maíta	Cuba	16	Male	18-25		LCA/Contact	A.D. 1250-1600[†]	Valcárcel Rojas et al., 2011
El Chorro de Maíta	Cuba	47	Male	26-35		LCA/Contact	A.D. 1250-1600[†]	Valcárcel Rojas et al., 2011
Canashito	Aruba	3	Unknown	adult		Preceramic	cal. 350 B.C. - A.D. 150	Versteeg et al., 1990; Wagenaar Hummelinck, 1959
Escape	St. Vincent	36	Male	36-45		ECA	A.D. 300-600[†]	Moravetz and Callaghan, 2011
Juan Dolio	Dominican Republic	10	Female	18-25		LCA	A.D. 900-1500[†]	Drusini et al., 1987; Veloz Maggiolo, 1972
Juan Dolio	Dominican Republic	22A	Male	18-25		LCA	A.D. 900-1500[†]	Drusini et al., 1987; Veloz Maggiolo, 1972
Kelbey's Ridge 2	Saba	132	Female	46+		LCA	cal. A.D. 1350-1450	Hoogland and Hofman, 1999; Weston, 2010
Kelbey's Ridge 2	Saba	313	Child	11-13		LCA	cal. A.D. 1350-1450	Hoogland and Hofman, 1999; Weston, 2010
Maisabel	Puerto Rico	2*	Male	26-35	cal. A.D. 670-1060	ECA/LCA	cal. A.D. 270-1150	Siegel, 1992; Weston and Schats, 2010
Maisabel	Puerto Rico	5	Female	adult	cal. A.D. 780-990	LCA	cal. A.D. 270-1150	Siegel, 1992; Weston and Schats, 2010
Maisabel	Puerto Rico	16*	Child	4-5	cal. A.D. 770-990	LCA	cal. A.D. 270-1150	Siegel, 1992; Weston and Schats, 2010
Malmok	Aruba	6	Unknown	26-35		Preceramic	A.D. 200-900[†]	Versteeg et al., 1990; Dijkhoff and Linville, 2004
Malmok	Aruba	10	Female	36-45		Preceramic	A.D. 200-900[†]	Versteeg et al., 1990; Dijkhoff and Linville, 2004
Manzanilla	Trinidad	118	Female	18-25		LCA	A.D. 400-1400[†]	Dorst, 2007; 2008; Jansen and Dorst, 2007; Weston pers. comm. 2010
Manzanilla	Trinidad	267/269	Male	26-35		ECA	A.D. 400-1400[†]	Dorst, 2007; 2008; Jansen and Dorst, 2007; Weston pers. comm. 2010
Manzanilla	Trinidad	291	Indet.	14-16		LCA	A.D. 400-1400[†]	Dorst, 2007; 2008; Jansen and Dorst, 2007; Weston pers. comm. 2010

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Point de Caille	Saint Lucia	32	Female	18-25		LCA	A.D. 1000-1400†	Friesinger et al., 1986
Point de Caille	Saint Lucia	36	Male	18-25		LCA	A.D. 1000-1400†	Friesinger et al., 1986
Punta Macao	Dominican Republic	6A	Female	18-25		LCA	A.D. 600-1500/1600†	Atiles, 2004; Tavarez Maria and Luna Calderón, 2007
Punta Macao	Dominican Republic	10A	Child	4-5		LCA	A.D. 600-1500/1600†	Atiles, 2004; Tavarez Maria and Luna Calderón, 2007
Punta Macao	Dominican Republic	11*	Male	26-35		LCA	A.D. 600-1500/1600†	Atiles, 2004; Tavarez Maria and Luna Calderón, 2007
Punta Macao	Dominican Republic	25	Female	26-35		LCA	A.D. 600-1500/1600†	Atiles, 2004; Tavarez Maria and Luna Calderón, 2007
Tanki Flip	Aruba	200	Unknown	adult		LCA	A.D. 950-1400†	Versteeg and Rostain, 1997
Tutu	St. Thomas, USVI	16	Female	36-45	cal. A.D. 640-870	ECA	cal. A.D. 65-950 / cal. A.D. 1150-1500	Righter, 2002; Sandford et al., 2002
Tutu	St. Thomas, USVI	30	Male	36-45	cal. A.D. 1300-1425	LCA	cal. A.D. 65-950 / cal. A.D. 1150-1500	Righter, 2002; Sandford et al., 2002
Tutu	St. Thomas, USVI	32A	Child	5		ECA?	cal. A.D. 65-950 / cal. A.D. 1150-1500	Righter, 2002; Sandford et al., 2002

Table 1.

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Table 2

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Site	AAG	AAG	AAG	CB	CM	CM	CS	ES	JD	JD	KR	KR	MB	MK	MK	MZ	MZ	MZ	PC	PC	PM	PM	PM	TF	TT	TT	TT
Burial	196	197	2217	85-40-17	16	47	3	36	10	22A	313	132	5	6	10	118	267/269	291	32	36	6A	10A	25	200	16	30	32A
Calathea sp.		1																									
Canavalia sp.		1										1															
cf. Cannaceae																											2
Fabaceae		1	1	1								1									1						1
cf. Fabaceae	1								1			1														1	1
Ipomoea batatas				1					1	1										1		2					
cf. *Ipomoea batatas*				1			2				1														1		
Leguminoseae						3		2																			
Manihot esculenta															1												
cf. Marantaceae				1																							
Phaseolus vulgaris									1																		
Phaseolus sp.										1																	
cf. *Sagittaria* sp.													7														
Xanthosoma cf. *sagittifolium*			1																								
Xanthosoma sp.														cl ~30													
cf. *Xanthosoma* sp.			1																								cl ~50
Zamia sp.						1											1	2									
cf. *Zamia* sp.							1									1	1	2						1			
cf. *Zamia erosa* (syn. amblyphyllidia)													2														
Zamia pumila				1																			2				
cf. *Zamia pumila*										2													1				
Zea mays	2				1		1		3				1			2	1		1				1	1		3	
cf. *Zea mays*							2								1	3	1		1						2	2	
Unidentified	1	1	2			1	6	2	1			1	4	6	1	4	2	3				1	2	2	3		5
Unidentified transitory starch																									cl ~13		2 cl ~35/50
Grain damage	P P/H	P P/H	P P/H	P P/H?	P/H	P P/H H?	P P/H H	P	P P/H H?	H P?	H	P	P P? P/H? H?	P	P H	P P/H P/H? H	P H/W	P P/H H	P P/H		P	P	P P/H P/F? H	P P/H? H	P P/H H? H	P P/H H	P P/H

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															H/ W												
Total*	**4**	**4**	**5**	**5**	**1**	**5**	**12**	**4**	**7**	**4**	**1**	**4**	**14**	**6**	**3**	**10**	**6**	**7**	**2**	**1**	**1**	**3**	**6**	**4**	**6**	**6**	**10**

Table 2.

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Table 3

Taxa*	**Size ranges in µm**	**Mean size in µm**	**Total no. of identified starches**
Domesticates			
Manihot esculenta	26.3	26.3	1
Phaseolus vulgaris	28.9	28.9	1
Zea mays	11.6 - 30	21.7 (±4.2)	17
cf. *Zea mays*	11.3 - 33	23.4 (±5.6)	10
Cultivars			
Ipomoea batatas	24.8 - 37.5	30.4 (±3.7)	6
cf. *Ipomoea batatas*	13.2 - 48.8	28.5 (±13.2)	5
Xanthosoma cf. *sagittifolium*	24.4	24.4	1
cf. *Xanthosoma* sp.	12	12	1
Wild			
Zamia pumila		34.8 (±5.7)	3
cf. *Zamia pumila*	26.3 - 43.1	43.8 (±24.5)	3
Zamia sp.	16.9 - 83	47.6 (±15.1)	4
cf. *Zamia* sp.	33.8 - 66.8	36.3 (±9.8)	6
cf. *Zamia erosa* (syn. *amblyphyllidia*)	18.8 - 45	27.6 (±7.2)	2
Canavalia sp.	22.5 - 32.6	41.3 (±14.6)	2
Calathea sp.	25.1 - 56.2	54.4	1
Fabaceae	54.4	27.7 (±11.3)	6
cf. Fabaceae	8 - 42	31 (±11.5)	5
Leguminoseae	11.3 - 46.9	31.4 (±9.1)	5
cf. *Sagittaria* sp.	11.3 - 45.8	24.6 (±5.1)	7
cf. Marantaceae	16.9 - 33.8	21.4	1
cf. Cannaceae	21.4	37 (±2)	2
	18.8 - 56.3		

Table 3.

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Figure 1

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Figure 2

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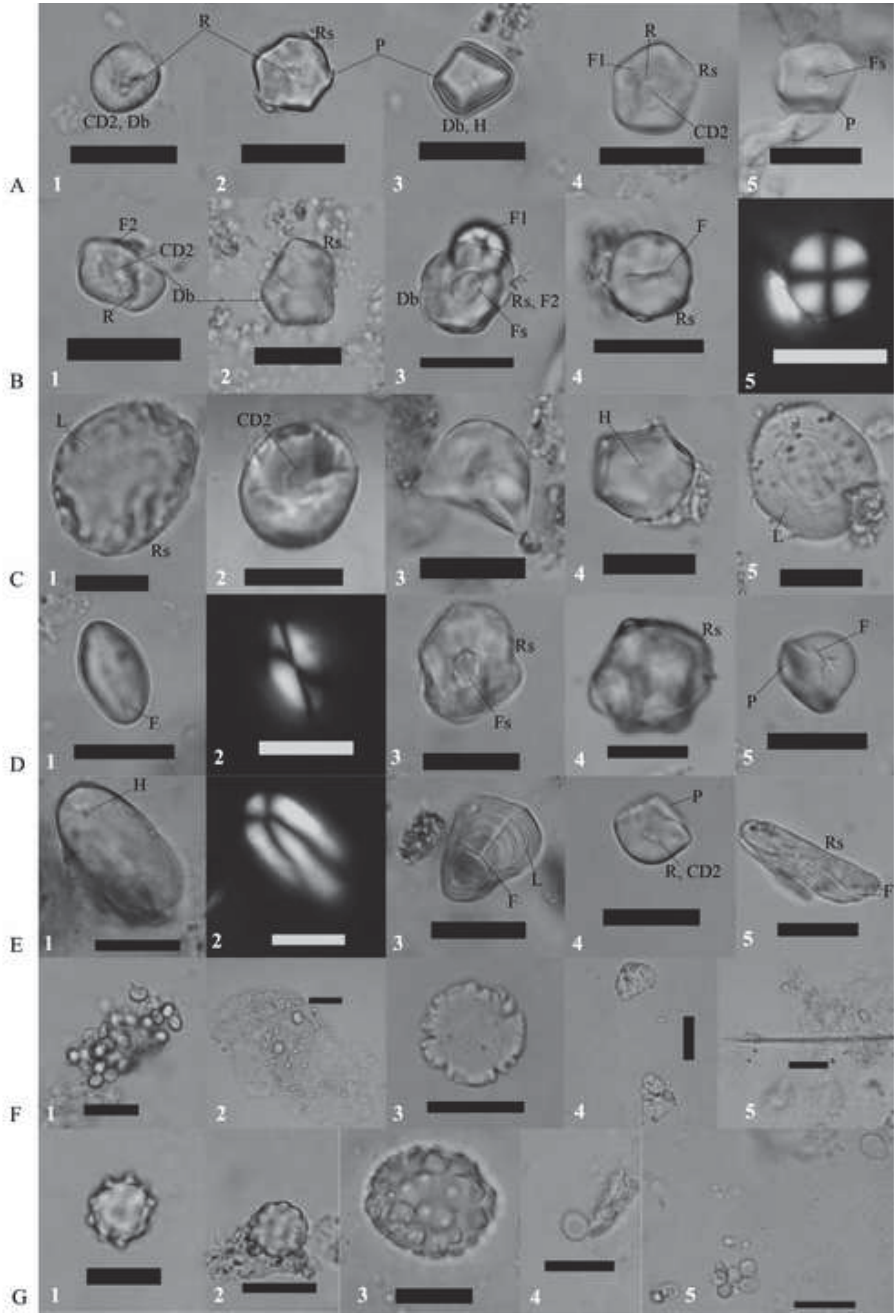

Figure 1 Appendix A

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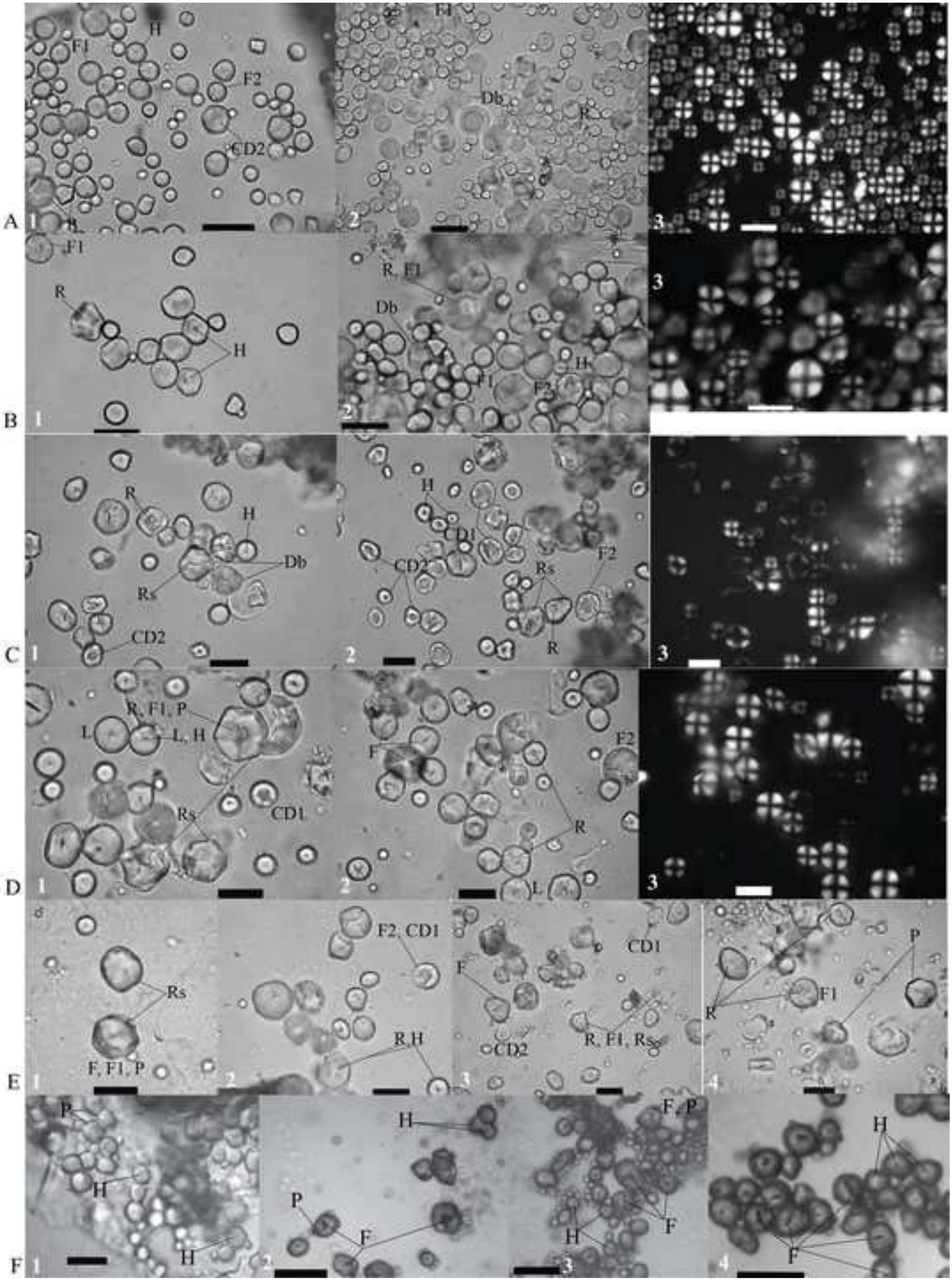

Appendix A - Grinding experiment with modern maize landraces

Introduction

The majority of the recovered archaeological starch grains (87.9%, see article Table 2) show variable degrees of damage, and variation in patterns of damage due to grinding. Maize starches in particular, while exhibiting many of the diagnostic features defined for the species, were found to be consistently larger than previously documented in modern and ancient maize starches. In order to assess this apparent discrepancy Pagán-Jiménez developed a single grinding experiment using two indigenous Caribbean maize landraces: Pollo (originally from Colombia/Venezuela) and Nal-Tel (originally from the Yucatan Peninsula in Mexico). These two races were selected for their diagnostic features previously defined for maize starch grains (e.g., Holst et al., 2007; Pagán-Jiménez, 2007; Pearsall et al., 2004). The results of this grinding experiment were compared to previous experiments by Pagán-Jiménez (2007). In this previous experiment, maize starches were extracted from the kernels after soaking them in water for 24 hours. The kernels were ground with a marble mortar and pestle, for no longer than 15 seconds in a liquid environment. Starches were allowed to settle in receptacles and finally filtered with small sieves to extract seed fragments. The results of this previous experiment are use here as control samples.

Materials and method

Seeds (kernels) of both maize landraces were acquired at the Seed Inspection and Distribution Unit in CIMMyT, Mexico. Pollo landrace, commonly regarded as primitive maize (Roberts et al., 1957), is known to be geographically distributed across both high and low altitudes in northern South America (Roberts et al., 1957; Sanoja, 1997). It produces small, short (~8.8 cm in length) and mostly conical ears with an average of 10 rows of seed. Kernels are short and rounded, and the endosperm is flinty (pop type) and rarely floury. Nal-Tel is a landrace from the lowland Yucatan Peninsula and low to middle altitudes in Guatemala where various sub-races grow (Wellhausen et al., 1952; Wellhausen et al., 1957). Similar to Pollo, Nal-Tel produces small, short (~12.1 cm in length) and mostly conical ears with an average of 12 rows of seeds. Kernels are rounded to dorsally flattened and sometimes slightly dented; the endosperm ranges from semi-hard to flinty. Both types mature rapidly and have a wide adaptation range (Wellhausen et al., 1957).

The premise of this experiment was the assumption that there is a direct relationship between endosperm hardness and kernel resistance to grinding, meaning that starches from kernels in different states of maturation and hardness should display different patterns of damage as they respond differently to pressure/grinding. In order to test this relationship, three different categories were defined for the maize kernels based on their maturity and hardness (a) green (soft kernels), (b) semi-mature (partially hard kernels), and (c) mature (hard and dry kernels). Previous observations regarding the relation between maize starch shape (spherical/round vs. polygonal) and endosperm hardness have been published elsewhere and do not form part of this experiment on damage by grinding (Cortella and Pochettino, 1994; Pagán-Jiménez et al., 2005). Kernels representing these categories were then subjected to intense grinding with a marble mortar.

Green, semi-mature, and mature kernels were selected from the Pollo landrace after growing maize plants from seeds from CIMMyT. Plants were grown in Pagán-Jiménez' experimental plot at Río Piedras, Puerto Rico, at an elevation of 20 mamsl. All samples were taken from the same ear at different developmental stages. Kernels were carefully extracted from the ear in order to avoid damaging the starches prior to grinding. Nal-Tel mature and dry kernels were selected from those delivered by CIMMyT. In total, four kernel groups (three Pollo and one Nal-Tel) were ascribed to the three main categories defined above (Table 1).

Three of the four kernel groups, excluding the green ones (Pollo), were soaked for one hour prior to grinding to avoid them “leaping” from the mortar during grinding. The soaking process softened the kernels very slightly, but only in the outer parts. The kernels were ground using a marble mortar and pestle. Each kernel group was ground separately, and all implements were thoroughly washed with sterile brushes and distilled water after processing each group. All kernels, at least ten in each group, were intensively ground continuously for five minutes in a liquid environment (2 ml of distilled water). A single drop of the resulting starchy solution was placed on a sterile microscope slide and a half drop of liquid glycerin was added and mixed to add viscosity. Individual starch grains were selected randomly; 30 were analyzed for each kernel group using an Olympus BH-2 microscope with polarization capacity at 40X. The starches were described according to previously defined morphometrical criteria (Pagán-Jiménez, 2007), but adding two new variables for surface topography: the presence or absence of central depressions, and the presence or absence of a bright ring associated with pressure around the hilum.

Results

While many statistical (e.g., multivariate) analyses can be made for describing starch grain morphometrics of the selected maize landraces, we focused on results derived from single descriptive statistics. We present here some general, but important statements on patterns of damage and their relation to starch grain morphometrics.

Shape

Modern maize starches from the four groups of kernels revealed highly variable shapes (Figure 1, Rows A-D). In all cases, irregularly oval/polygonal shapes (between 9 to 14 different variants) were registered alongside regular shapes such as spherical/round to oval in each group (Table 1). In the Pollo semi-mature kernels, the vast majority of the starches were irregular and highly variable (14 different variants were recorded) in shape. In the control samples, an “intermediate” proportion between irregularly oval/polygonal and spherical to oval shapes was registered (i.e., 76.2% vs. 23.8%) in Pollo starch grains (Pagán-Jiménez, 2007: 250; Figure 1, F2). In the control samples the number of shape variants was considerably higher (up to 30 different variants). These results indicate that more intensive grinding of the kernels at the different developmental stages does not produce greater variety in maize starch shapes. Comparison with the control samples and previously published results does suggest, however, that during the process of intensive grinding, damage to the starch grains tends to somewhat homogenize starch shapes, as the proportion of spherical to oval starch shapes increases in comparison to the irregular oval and polygonal shapes (cf. Holst et al., 2007; Pagán-Jiménez, 2007; Table 1). Thus, we may expect that archaeological maize kernels submitted to intensive grinding will yield a greater proportion of "regular", but identifiable, starch grains than those typically thought as characteristics of the genus (e.g., irregular or polygonal shapes).

<<Figure 1>>

Modern maize starch grains from the grinding experiment. Rows A–C: Pollo. Row D: Nal-Tel. Row A: starches of green (soft) kernels. Row B: starches of semi-mature (partially hard) kernels. Row C: starches of mature (dry and hard) kernels. Row D: starches of mature (dry and hard) kernels. Row E: Images of modern starch grains from other maize landraces which were also soaked for one hour and intensively ground for five minutes are included for visual comparison, starches of mature (dry and hard) kernels: (E1) Chapalote; (E2) Confite morocho; (E3, E4) Pira. Row F: Images of modern starch grains of mature (dry and hard) kernels from control samples (Pagán-Jiménez, 2007): (F1) Starches of Pollo mature kernels as seen directly in the endosperm (without grinding, this study); (F2) Starches of Pollo; (F3) Starches of Early Caribbean; (F4) Starches of Negrito de Colombia. CD1, large central depression; CD2, small central depression; R, bright ring; Db, double-border (double-outline edge); F, general fissures; F1, radial fissures or striations; F2, radial to asymmetric fissures; P, pressure facets; H, hilum; L, lamellae; Rs, rough topography. All scale bars are 30µm. Dark field micrographs with cross polarized light show general Maltese crosses and correspond to the same starches in the images immediately to the left.

Maize landraces	**Spherical to oval (%)**	**Irregularly oval to polygonal (%)**
Nal-Tel (mature, dry and hard)	46.7	53.3
Pollo (green and soft)	46.6	53.4
Pollo (semi-mature and partially dry)	13.3	86.7
Pollo (mature, dry and hard)	30	70

Table 1. Proportion of regular vs. irregular shapes in modern maize starch grains per kernel group as observed during this experiment.

Size

The size ranges documented in this experiment (Table 2) are revealing when contrasted with previously published data (e.g., Holst et al., 2007; Pearsall et al., 2004). Nal-Tel mature and Pollo semi-mature kernels are between 6 – 13 µm bigger than control samples of Pollo mature kernels (Pagán-Jiménez, 2007; Figure 1, compare Rows A-D with Row F). This difference in size range can be attributed to the enlargement of the starch grains due to the grinding process as the starches previously studied by Pagán-Jiménez (2007) were not subjected to such a long period of intensive grinding as is the case in the present experiment. Three of the four groups of kernels (Table 2) showed size ranges (mean sizes between 20.8 – 23.2 µm) above the norm when compared to previously known maize starch size ranges (Holst et al., 2007; Pagán-Jiménez, 2007). The mean size measured for Pollo green kernels (12.1 µm) falls within previously known size ranges.

These data suggest that there is a relationship between substantial enlargement of starch grains and the degree of hardness of the maize kernel from which they are derived: the harder the kernel, the greater the enlargement of the starches during intense grinding.

***Zea mays* (modern samples)**	**Range of sizes in µm**	**Mean sizes in µm.**	**No. measurements taken**
Control samples (Pagán-Jiménez, 2007) Mature, dry and hard kernels soaked for 24 hours before grinding [1]			
a. *Pollo* (CIMMyT Id#:3106)	2-28	13 (±3.9)	116
b. *Early Caribbean* (CIMMyT Id#:1347)	3-20	13 (±3.6)	101
c. *Negrito de Colombia* (CIMMyT Id#:3199)	5-20	12.3 (±3.3)	107
d. *Cateto cristalino* (CIMMyT	3-18	10.3 (±3.1)	107

Id#:4113)			
e. *Chandelle* (CIMMyT Id#:3879)	2-20	12.3 (±3.2)	89
f. *Tuśon* (CIMMyT Id#:5495)	1-18	12 (±3.2)	109
Experiment samples (this study) Green to dry kernels soaked, or not (marked with * below), for 1 hour before grinding[2]			
g. *Nal-Tel*,mature, dry and hard (CIMMyT Id#: 815)	11-41	21.8 (±7.7)	60
h. *Pollo*, mature, dry and hard (CIMMyT Id#: 3105)	10-38	23.2 (±6.6)	60
i. *Pollo*, semi-mature and partially hard (CIMMyT Id#: 3105)	7-34	20.8 (±5.7)	60
j. *Pollo**, green and soft (CIMMyT Id#: 3105)	5-25	12.1 (±4.7)	60

Table 2. Modern starch grain size ranges of seven indigenous landraces submitted to different grinding processing and kernel conditions.

1. After soaking, kernels of Control samples were ground for 15 seconds with a marble mortar and pestle to avoid overly damaging the starch grains (Pagán-Jiménez, 2007).

2. After soaking, kernels of Experiment samples were ground intensively for five minutes with a marble mortar and pestle in order to examine patterns of damage due to pressure.

Fissures and striations

Fissures and striations are produced naturally during development of the kernel, as the seed looses water and hardens, directly affecting starch packages within amyloplasts. It is known that fissures and striations are also often the result of mechanical processes such as grinding and sometimes heating (parching), and fermentation (Dorsey et al., 2009; Pagán-Jiménez, 2007). The grinding experiment revealed that Pollo green kernels were the least affected by thin radial fissures, with just 6.3% of the starch grains affected. The other kernel groups were affected far more frequently by fissures, with radial fissures being the most commonly observed type (Figures 1 and 2). Again this seems to indicate a relation between the hardness of the kernel and the amount of damage caused during intense grinding, thus suggesting that these fissures were the result of human action (grinding). Figure 2 also shows the relation between harder kernel types and larger starch grains, with the tendency for enlargement of the starches in all types but the green Pollo variant reflected across the categories of fissures.

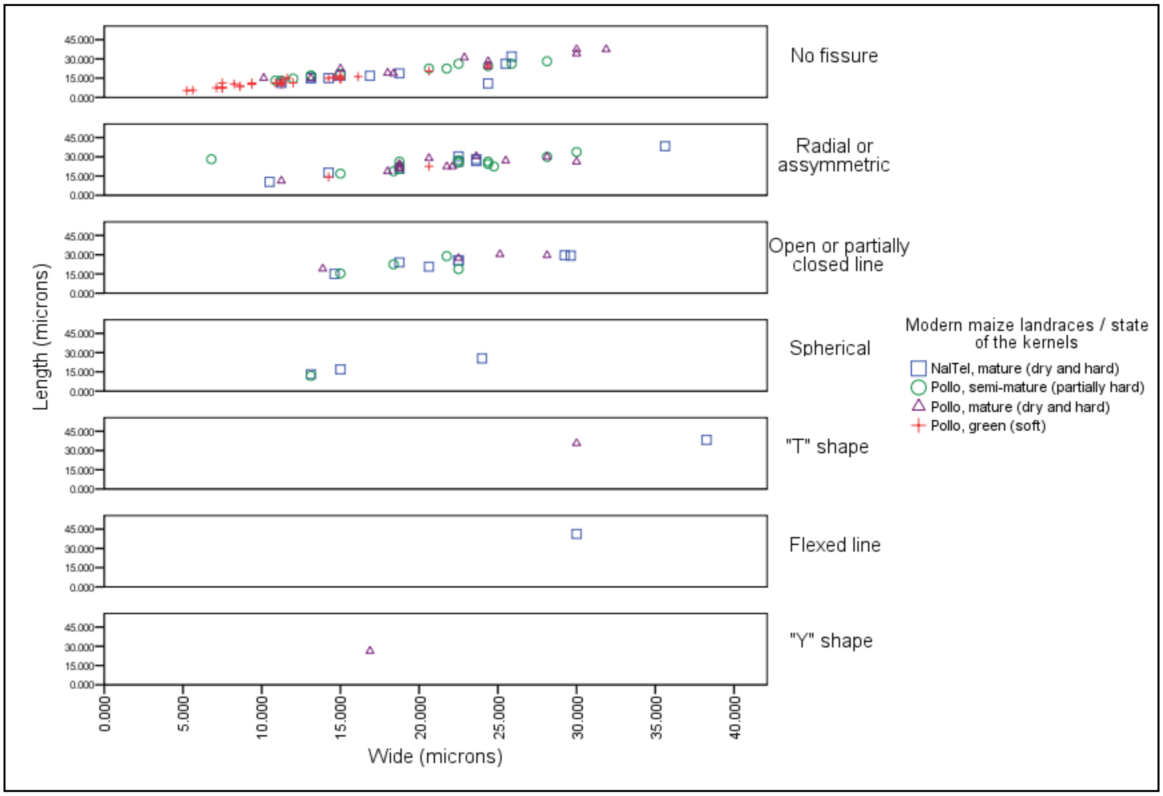

Figure 2. Size by fissure variants among modern maize starch grains.

Surface topography

Some previous studies have defined surface (i.e., topographic) characteristics of maize starch grains (e.g., Pearsall et al., 2004; Dorsey et al., 2009). Here, the assessment of the grain surface focused on distinguishing surface alterations potentially caused by pressure (intense grinding). A distinction made based on the on the appearance of the starch grain surface: rough (i.e., irregular topography) or normal (i.e., smooth topography). In all four of the kernel groups smooth surfaces were the most commonly registered type (73.3%; Figure 3). The remaining 26.7% consisted of rough surface starches from the Pollo semi-mature, and mature kernels, and from the Nal-Tel mature kernels (see article Figure 1). None of the green Pollo kernels displayed rough surfaces. According to these results it is reasonable to suggest that intense grinding of hard kernels (e.g., Pollo and Nal-Tel mature) results in the increased roughness of starch grain surfaces, while the same mechanical process in softer kernels will either minimally affect the surfaces (e.g., Pollo semi-mature), or will not affect them at all (e.g., Pollo green kernels). Figure 3 furthermore shows that rough surfaced starches generally show a greater tendency for enlargement during the grinding process than smooth surfaced starches, possibly pointing toward a relation between these two effects of pressure/grinding.

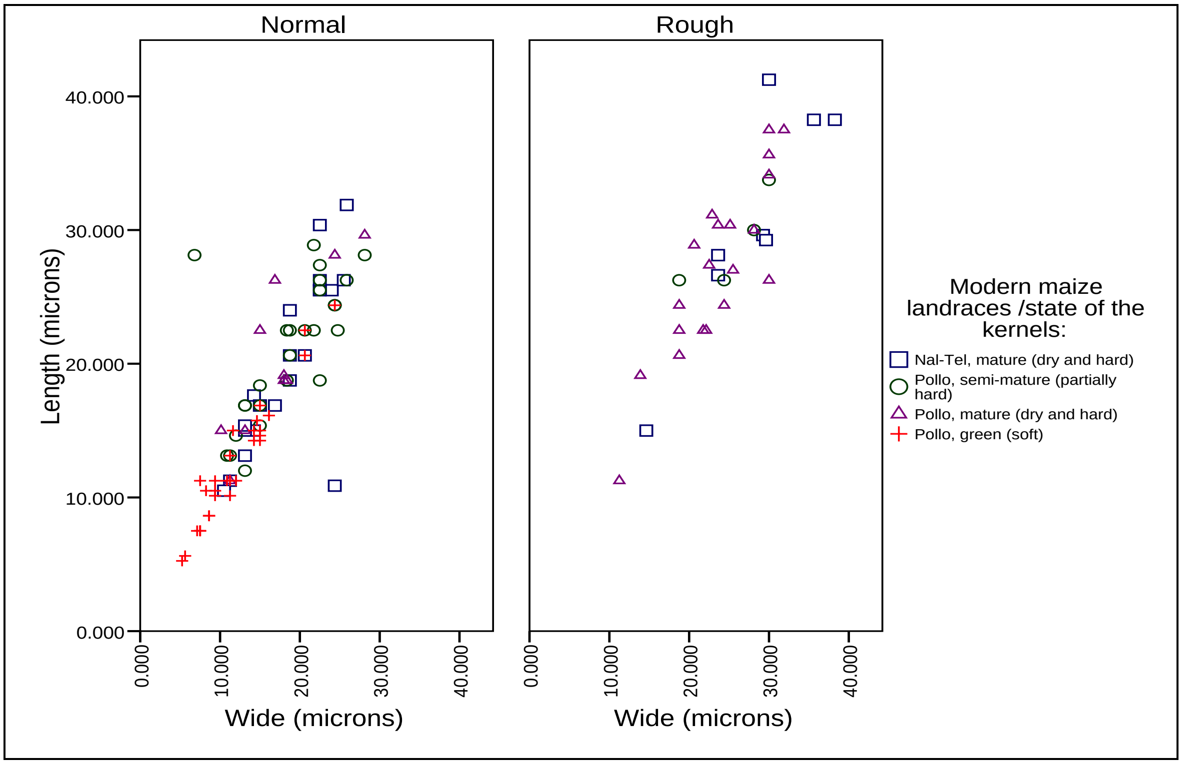

Figure 3. Size by surface topography among modern maize starch grains.

Central depressions

Central depressions have been described elsewhere as scooped features created during grinding (Dorsey et al., 2009) or as large circular depressions (known as “folds”, “shadows”, or depressed circles) resulting from boiling of the source organs of the plants for 1 – 10 minutes (Henry et al., 2009). To assess whether central depressions may also be related to pressure damage, we registered the presence or absence of central depressions in the starches from the four kernel groups, identifying two variants: (1) a small to medium size central depression commonly found in the hilum area, (2) a large central depression that extends to the edges of the grain (Figure 1, Rows A-D). Both variants were grouped together as a single feature (central depression) (Figure 4), which was observed in 26.7% of the starches of the four kernel groups. Similar to other morphometric features described above, the prevalence of central depressions tends to increase with greater kernel hardness and resistance to intense grinding. Central depressions were observed in only 20% of the Pollo green kernels, compared to 40% and 53% in Nal-Tel and Pollo mature kernels respectively. In Figure 4 we see that starches with a central depression generally show a slightly greater tendency for grain enlargement than starches without central depressions, perhaps indicating a relation between these two effects of pressure/grinding.

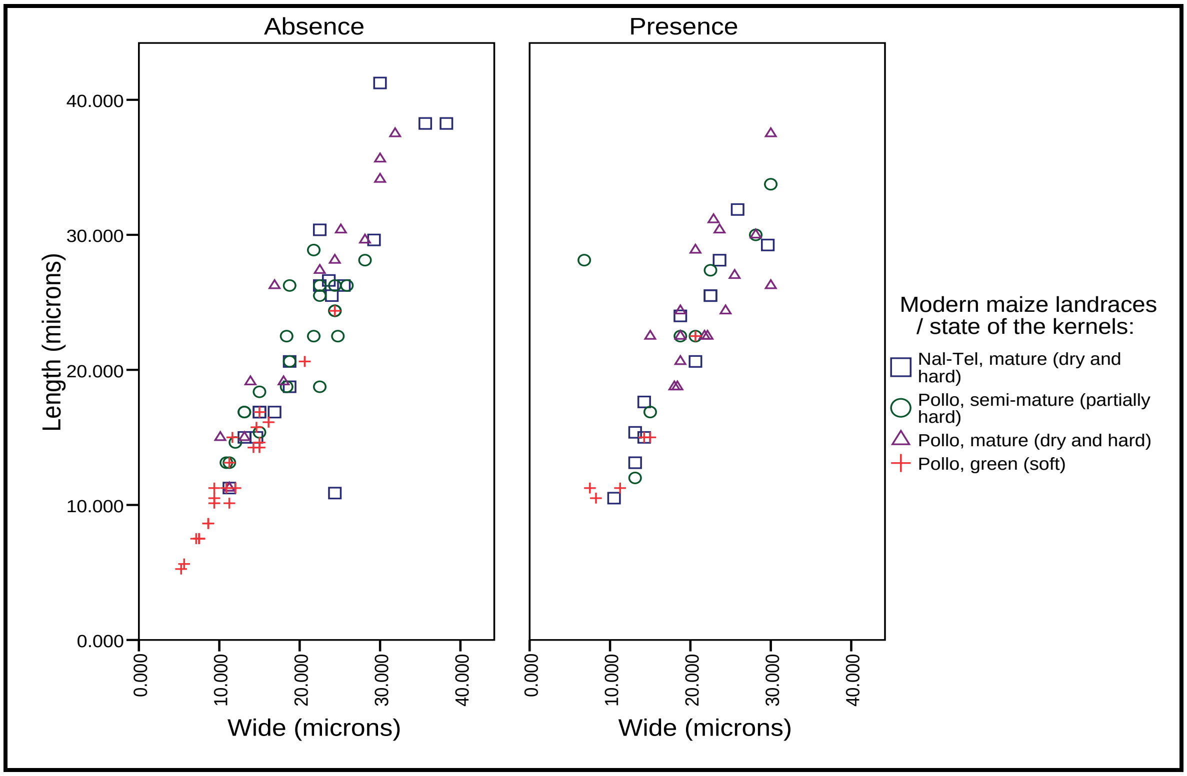

Figure 4. Size by central depression among modern maize starch grains.

Bright ring around hilum

The last morphometric feature assessed here is the presence of a single bright ring around the hilum (Figure 1, Rows A-D). To the best of our knowledge, this feature has not previously been described by other researchers as resulting from grinding damage or other mechanical processes, however we have observed the bright ring only in maize starch grains submitted to intense grinding.

Similar to the other pressure/grinding features assessed in the grinding experiment, the bright ring was registered on a presence/absence basis (Figure 5). Results show that this feature was present in 25.9% of starches of the four kernel groups. Pollo mature kernels were most frequently affected (36.6%), while in the Pollo green type it was observed in only 6.6% of starches. Of the Nal-Tel mature kernels starches, 26.6% displayed the bright ring; of the Pollo semi-mature, 33% of the starches were affected. Again this feature is possibly a diagnostic feature of intense grinding damage in maize, as there is a greater tendency for harder kernel types to present a bright ring than softer kernel types. Figure 5 illustrates a slight correlation between the presence of a bright ring around the hilum and grain enlargement, again showing these two effects of pressure/grinding may be related.

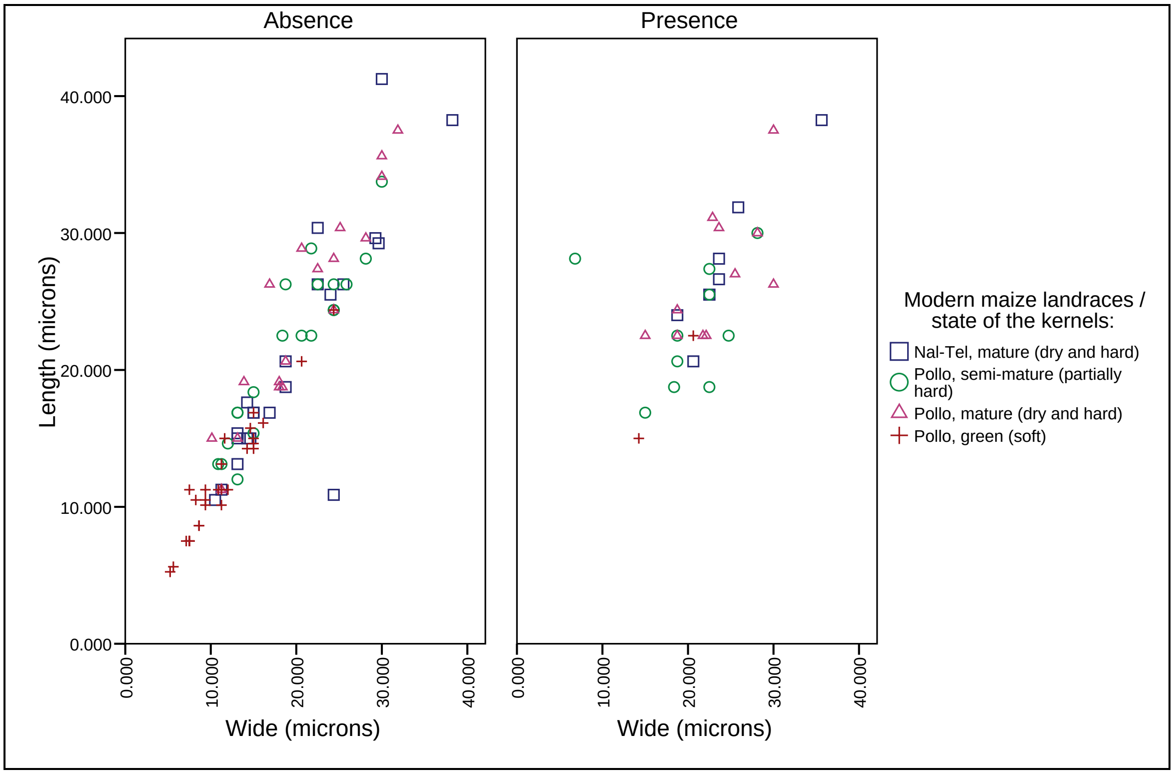

Figure 5. Size by bright ring among modern maize starch grains

Conclusions

The results of this experiment appear to support the hypothesis that starch stored in the different maize kernel groups will display differential damaging after intense grinding as the reaction to pressure/grinding is determined by matrix hardness. Based on these results we propose that there is a direct relationship between substantial enlargement of maize starch grains and the degree of hardness of the maize kernel from which they are derived; or in other words the harder the kernel, the greater the enlargement of the starches during intense grinding. And vice versa is true: in softer kernels the enlargement is minimal.

Furthermore, other damage features such as fissures and striations, altered surface topography, and central depressions appear to be related to pressure damage during grinding of the maize kernels. In sum, there is a clear relation between the prevalence of these damage patterns and kernel hardness; the harder the kernel, the more frequently these damage patterns are observed after intense grinding. Our observations of the presence of a bright ring around the hilum – and, similar to the other damage patterns, its relation to kernel hardness and intense grinding – mean a new pattern of pressure/grinding damage can now be distinguished.

These results also affect our understanding of characteristics previously considered to be diagnostic morphological features of maize starch grains (i.e., hilum, pressure facets, and a double-border [or double-outline edges according to Berman and Pearsall, 2008]), as these may now be considered to be the result of (or at least affected by) intense pressure/grinding. In these cases, the harder the kernel is, the less visible (but not necessary absent) the hilum, pressure facets, and external double-border become after intense grinding (Figure 1).

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Appendix B - Notes on the identification and diagnostic characteristics of *Zamia* sp., *Ipomoea batatas*, *Xanthosoma* sp., *Sagittaria* sp., *Manihot esculenta*, Cannaceae, Marantaceae, Leguminoseae, Fabaceae, and *Zea mays*

Zamia sp.

The identification of *Zamia* or marunguey in this study (see article Table 2; Figure 2, Row C) is based on a combination of observed features in the recovered starches: shape (spherical to polygonal), size ranges, the presence of ubiquitous radial or asymmetrical fissures, the presence of and generally unusual location of the hilum, the projection of lamellae, and the projection of a radiant Maltese cross with one or two 'wavy arms'. Starches produced in the tuberous stems of three current Caribbean species of marunguey, *Zamia pumila, Z. erosa* and *Z. portoricensis,* as well as other species found in Cuba (Roberto Rodríguez pers. comm., 2007), are thus far the only known starches in the insular Caribbean that alternate between oval, spherical to polygonal shapes and fall within a size range of 1 - 95µm. Common size ranges of these species are between 4 - 46µm (compare with recovered *Zamia* starches in article Table 3). To date, modern starches of other insular or continental Caribbean plants studied by us, or described elsewhere, do not fall within this size range. Nor do they display other distinctive features such as the spherical to polygonal shapes, marked symmetric lamellae and radial or asymmetrical fissures.

Differentiation among species in this study relies on the currently known geographic distribution of *Zamia* at regional (inter-island) and local (intra-island) levels, together with data on starch shape and size ranges (Table 3). At present, several wild populations of *Zamia* are known in Cuba, Hispaniola (Haiti and the Dominican Republic), Jamaica, Puerto Rico, the Bahamas, and the Cayman Islands. There is no evidence for its natural distribution throughout the Lesser Antilles or Trinidad and Aruba at any point in the past or present (Stevenson et al., 2003). There appears to be a direct relation between *Zamia* distribution and geological formations such as limestone, serpentine and/or quartzite. *Zamia pumila* is the only known species in the Dominican Republic, occurring in areas with limestone-karstic formations. The three archaeological sites in the Dominican Republic studied here are located in such areas. In Cuba, *Zamia pumila*, *Zamia erosa* and

Zamia ottonis are also distributed along serpentine or limestone formations in the east and in the province of Holguín, where the site of Chorro de Maíta is located (González Géigel, 2003). In Puerto Rico, three species of *Zamia* are currently known (Pagán-Jiménez, 2007). In the northern limestone-karstic region where the site of Maisabel is located, only *Zamia erosa* is found. The other two species (*Zamia pumila* and *Zamia portoricensis*) are distributed across the limestone and serpentine areas of the south and southwestern part of the island. The latter are geographically separated from *Zamia erosa* by the Cordillera Central.

Zamia sp. identifications for Lesser Antillean sites and individuals could not be confidently made to a higher resolution, due to the lack of evidence for its past and/or present natural distribution in the area. Starches attributed to *Zamia* in these sites fall within the accepted size range of modern species, and some of them also display the typical Maltese cross with at least two ‘wavy arms’, while others display an irregular hilum. The morphometric characteristics of one of the starches ascribed to *Zamia* in Manzanilla, Trinidad (see article Figure 2, C4), have been documented only in modern *Zamia pumila* and *Zamia portoricensis* (Pagán-Jiménez, 2007). Some islands in the region, such as Aruba, Guadeloupe, and Trinidad are characterized by important limestone and volcanic formations that could easily support natural populations of several Neotropical *Zamia* species, such as those known for the other islands in the Caribbean and areas of the adjacent continental landmasses. Further paleoethnobotanical research in these areas will clarify current issues of identification of this genus.

Ipomoea batatas

Secure and tentative identifications of sweet potato were based on distinctive and sometimes diagnostic features previously defined for the species (Pagán-Jiménez, 2007; Piperno and Holst, 1998; Reichert, 1913; see also Perry, 2002). Recovered starches displayed mostly polygonal shapes and many of them also showed two or three diagnostic pressure facets. Different types of fissures (from single to “Y”-shape lines) were common, while thin striations related to pressure damaging were also observed. The hilum (sometimes open) was often observed to be in an unusual position. The maltese cross was also commonly observed and frequently displayed narrow arms. In some cases this feature was unclear, probably due to pressure or heat damaging. Another distinctive feature was the projection of lamellae in which rings appear to follow the outline of the body. Size ranges for tentatively and securely identified sweet potato starches are 28.5 ±13.2µm and 30.4 ±3.7µm respectively (see Table 3), falling within previously documented ranges. In some cases, larger sizes were documented, beyond the expected size range for the species. In these cases the starch grains presented with clear signs of pressure damage (possibly grinding), heat (boiling), or both (Figure 2, D3-D4), all of which could promote starch enlargement (Babot, 2003; Henry et al., 2009; see also Appendix A) while retaining other distinctive features of the species such as shape and pressure facets. In short, previously described distinctive features were documented, with secure identifications being based on the presence of the majority (or in some cases all) of these features, and tentative identifications based on the presence some of these features.

Xanthosoma sp.

Secure and tentative identifications of cocoyam starches to the genus or species level (see article Tables 2 and 3) were based on the presence of previously described distinctive features of the taxa, such as the spherical and hemispherical to polygonal shapes in tandem with one to three pressure facets. Sizes range between 12-24µm in isolated starches which is consistent with white cocoyam (*Xanthosoma sagittifolium*), while clustered starches found in two different cellulosic tissues ranges between 0.4-15µm, which are closer to other cocoyam species such as *Xanthosoma undipes* (see Appendix B in Pagán-Jiménez, 2007). A more precise identification of the isolated starches could not be achieved due to patterns of damage (thin striations and a central depression) consistent with pressure (grinding or pounding) that could have caused starch enlargement or distortion of some distinctive morphological features. Starch clusters either securely or tentatively identified to the genus level were impossible to analyze in greater detail, as they could not be rotated to document all morphometrical features.

Sagittaria sp.

In an individual from the site of Maisabel, Puerto Rico, starches were recovered that match closely with many of the diagnostic features described for rhizomatous arrowhead

(*Sagittaria lancifolia*). The shapes of modern arrowhead starches are mostly spherical to oval with distinct undulation of the margins. Fissures are frequently observed, and occur as single or a number of asymmetric lines near the hilum area, which is mostly open and irregularly shaped. One of the most distinctive features of these modern starches is their heavily pronounced lamellae and the symmetric way these "wavy" rings follow the margins of the body. This latter feature, together with the spherical to oval shapes, the irregularly shaped hilum and size ranges between between 11 - 79µm (with a mean size of 54 ±8µm) appears to be diagnostic for the taxa, or at least to the arrowhead species. The recovered starches in this study, which are tentatively ascribed to *Sagittaria*, display all these features, with the exception of the size range, which is quite different (see article Table 3). We currently have no starches of other species of arrowhead native to tropical America in our reference collection, and descriptions of these species are lacking in the literature. Therefore, we are inclined to maintain this tentative identification to the genus level on the basis that some other genera (e.g., *Xanthosoma*, *Zamia*, *Phaseolus*) also show and share diagnostic characteristics within its many species, while displaying deviant size ranges.

Manihot esculenta

Manioc was identified in this study by a single starch grain found in individual 10 from the site of Malmok, Aruba (see article Table 2). Its shape is hemispherical (or bell-shaped) with a notable stellate fissure located in the hilum area, which is lightly irregularly shaped. Two concave pressure facets are clearly present in the basal end of the starch and the overall surface is notably smooth (Figure 2, D5). These features have all been considered as diagnostic for this species (Duncan et al., 2009; Piperno, 2006; Piperno and Holst, 1998; Reichert, 1913). The recovered manioc starch grain is 24 x 26.3µm, which deviates somewhat from some ranges published for modern manioc starches. The common size range of modern manioc starches has been defined at between 5 - 20µm with a mean size of 13.5 ±3.7µm (Pagán-Jiménez, 2007), or around 15µm (Reichert, 1913). Nonetheless, Piperno (2006) and Duncan et al. (2009) has documented a broader size range for this species (6 - 28µm) that fits well with our findings. In a study of manioc starch grains, Perry (2002) has suggested that some factors could be influencing the apparent variability in sizes between different manioc specimens: (a) root size (larger roots yield larger starches), (b) soil fertility (low soil fertility leads to larger starches), and (c) unknown factors such as moisture, cultivation practices, cultivar specifics and the amount of time that a root was allowed to remain in the ground accumulating starch. So it is reasonable to suggest that any of these factors could have influenced the size of the manioc starch from Malmok.

Cannaceae

Two starches, tentatively ascribed to Cannaceae, were recovered from the calculus of an individual from the site of Tutu, St. Thomas (see article Table 2). Both starches appear to display the elliptical to ovoid shapes, and size range (53 x 18.8µm) documented in modern starches of the edible *Canna indica* as well as in *C. glauca* and *C. jaegeriana* (see article Figure 2, E5; see also Pagán-Jiménez, 2007 and Reichert, 1913). In one of the recovered starches, an irregularly shaped hilum, and surrounding lamellae radiating from the hilum area to the outer edges of the grain were documented. Both starch grains displayed rough surfaces and signs of pressure damaging (longitudinal fissures), affecting the visibility of the diagnostic features used for identification. These starches appear similar to those produced by some *Calathea* sp. in Central America (Dickau et al., 2007; Piperno and Holst, 1998), however based on Pagán-Jiménez' reference collection of Cannaceae and *Calathea* starches from plants distributed throughout the insular Caribbean, these starches appear most similar to those produced by Cannaceae.

Marantaceae

A single Marantaceae starch grain was tentatively identified by its oval shape with slightly wavy margins, the eccentric location of the hilum and the projection of the Maltese cross. These features, however, are shared by different species of Marantaceae such as *Calathea* sp. and *Maranta* sp. The size of this starch grain (21 x 13µm), is also characteristic for these different species.

Another oval starch grain, displaying lamellae, a hilum and a Maltese cross very similar to those in some *Calathea* sp. (Figure 2, E1 and E2) was identified. These diagnostic features observed in this starch grain are distinctive to a number of species in the genus, but are most common in the tuberous roots of the South American *C. rufibarba* (see article Table 2), a species of not known to have been used by humans .

Other plants of the Zingiberales order (e.g., Heliconiaceae or Zingiberaceae) to which Marantaceae also belong, produce starches very similar in appearance to the one ascribed here to *Calathea* sp. Based on observations of Marantaceae starches in our reference collection, an important difference has been observed: lamellae in Marantaceae starches consist of well-defined and symmetrically curved arcs, while in species such as *Heliconia caribaea* the symmetrically curved arcs each display two marked obtuse angles. Further collection and study of starches from more specimens of the Zingiberales order from the insular Caribbean is needed to better understanding the diagnostics characteristics of these different species.

Leguminoseae and Fabaceae

A number of starches were tentatively or securely ascribed to different taxonomic levels of the Leguminoseae and Fabaceae families (*Canavalia* sp., *Phaseolus* sp., and *P. vulgaris*). Modern *Phaseolus vulgaris* and wild *P. vulgaris* starches have been described as simple oval to kidney-shaped grains with a centric hilum that usually has a distinct longitudinal fissure (Piperno and Dillehay, 2008). The starch size range of wild and domesticated specimens is 12 - 60µm. Generally speaking, *Canavalia* starches display very similar or identical diagnostic features to starches from many *Phaseolus* sp., although some minor differences have been suggested. In *Canavalia* starches, shapes are more irregular (Piperno and Dillehay, 2008), and fissures are less prominent in some of the wild species such as *C. nitida* and *C. rosea* (Appendix B in Pagán-Jiménez, 2007) in comparison to *C. plagiosperma* or *Phaseolus vulgaris/lunatus*. The starches recovered in this study display some of the main features described above. *Phaseolus* sp. and *P. vulgaris* starches were found in two individuals from the site of Juan Dolio (see article Table 2) and the observed diagnostic features of shape, lamellae, hilum and size range (see article Table 3) are consistent with those known for the genus. In one case a longitudinal (but smooth) fissure was observed (see article Figure 2, D1 and D2), while in another case a radiant ring around the hilum was observed, as well as possible pressure damage (grinding) in the form of a fissure perpendicular to the margins (Babot, 2003).

Two starches that closely match the features described for *Canavalia* sp. were also recovered in two individuals (see article Table 2; Pagán-Jiménez, 2007; Piperno and

Dillehay, 2008). However, in the absence of the (longitudinal) fissures documented in modern specimens, and considered typical for the species, identifications remain tentative.

In many other cases, while oval (but wavy) shapes and expected size ranges were observed, patterns of damage, most likely due to pressure and/or heat treatment, meant clear observation of some of the other important diagnostics features was not possible.

Zea mays

Both secure and tentative identifications of maize starch grains were made (see article Table 2 and Figure 2, Rows A and B). Different to other regions, where wild relatives of maize produce similar, but clearly distinguishable starches (Holst et al., 2007; Piperno et al., 2009), in the insular Caribbean no currently known plants, including wild grasses, possess starches comparable to maize starches. Maize starches were identified based on previously published diagnostic features (e.g., Holst et al., 2007; Pagán-Jiménez, 2007; Pearsall et al., 2004; Piperno and Holst, 1998). Shapes are spherical to polygonal and display multiple pressure facets according to matrix (endosperm) hardness (see Appendix A). The hilum is commonly open, slightly irregular, and most common in soft endosperm maize kernels. Lamellae are generally absent, and fissures with different patterning (“Y”, “T”, and triangular shapes) are common, especially in starches with dried or harder (flint) kernels. One of the main diagnostic features defined for maize starch is the presence of a double-border. Size is slightly variable among the different maize landraces (Holst et al., 2007; Pearsall et al., 2004) although a range of 2 - 28µm, with a mean size of 12.5 ±3µm for 7 indigenous landraces have been documented for our reference collection (Pagán-Jiménez, 2007).

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