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Changes in starch grain morphologies from cooking

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A B S T R A C T

One of the three main plant microfossils, starch grains are increasingly used as markers of diet, plant domestication, tool use and site organization, because their morphology and features provide a means to identify the plant that produced them. However, starch grains are susceptible to damage when they are exposed to heat in the presence of water, as in cooking. We documented the changes that occur in the starch grains of 10 domesticated plant species due to exposure to different cooking methods, in order to better understand how cooking alters the appearance of the grains, and if these cooking methods might be identifiable in the archaeological record. Our results show that some cooking methods produce unique, identifiable damage on some types of plant starches, but generally each plant species reacts uniquely to cooking. In order to record the changes for each plant species, we have created a database, available at (http://www.osresearch.net/~hollyf/starchdb/index.cgi), to which registered users can add their own images of cooked starch grains.

1. Introduction

Plant starch grains are increasingly being used by researchers as a means for recovering information about plants in the archaeological record. Starch grains, along with phytoliths and pollen, are plant microfossils, which are microscopic plant remains with unique morphologies that allow them to be identified in most cases to the family, genus or species of plant that produced them. Starch grains have been recovered from soils (Balme and Beck, 2002; Henry et al., 2004), stone tools dating from the Pleistocene through the Holocene periods (Barton, 2007; Fullagar et al., 2006; Pearsall et al., 2004; Piperno and Holst, 1998; Zarrillo et al., 2008) pottery (Crowther, 2005), and dental calculus (Boyadjian et al., 2007; Henry and Piperno, 2008; Piperno and Dillehay, 2008), and have been used to document diets of ancient animal and human groups, including plant domestication (Barton, 2007; Fullagar et al., 1996, 2006; Loy et al., 1992; Pearsall et al., 2004; Perry et al., 2007; Piperno et al., 2000, 2004; Zarrillo and Kooyman, 2006), and even social and spatial organization of sites (Balme and Beck, 2002; Henry et al., 2004).

Starch grains are energy storage bodies of plants, and are made of two types of long glucose chains, amylose and amylopectin. Plants may produce these starches in almost every tissue, but they are particularly abundant in tissues where long term energy storage is needed, such as in fruits, seeds and underground storage organs (Torrence and Barton, 2006). Starches can be identified in most cases as coming from a particular plant family, genus or species based on their size, shape and the presence and appearance of certain physical features, like hila (the nucleus around which the starch forms), lamellae (ring-like shadows of the boundaries between crystalline and amorphous layers of the starch), dimples, cracks and fissures (Cortella and Pochettino, 1994; Fullagar et al., 1996, 2006; Holst et al., 2007; Loy et al., 1992; Pearsall et al., 2004; Piperno and Holst, 1998; Piperno et al., 2000, 2004; Reichert, 1913, 1919; Seidemann, 1966; Torrence and Barton, 2006). In archaeology, starch grains are usually examined and identified using transmitted cross-polarized light microscopy at moderate magnifications, usually 200–400 times, and this has proven to be a time- and cost-efficient method. It also allows for a simultaneous examination of all three plant microfossils, although processing techniques developed specifically for each type of microfossil are necessary for a robust examination and quantification of each one (Piperno, 2006).

Unlike pollen and phytoliths, starch grains are composed of soft, relatively fragile organic material and are desirable energy sources for potential consumers. Because of these properties, they can undergo several pre- and post-depositional changes, such as from milling, grinding, cooking, parching and drying, and from soil borne enzymes and other microscopic consumers, like yeast (Lineback and Wongsrikasem, 1980; del Pilar Babot, 2003). These processes

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have the potential to damage or destroy the unique features of the starch grains. Much of the research on chemical and physical changes of starch grains during cooking or processing has been performed in the food science realm, where methods such as X-ray diffraction and SEM imaging are used to describe the changes to the starches. These studies have shown that heating starch grains in the presence of water breaks down the crystalline layers, and cause the starches to gelatinize, or form a viscous complex with water (McGee, 1984).

There are only a handful of studies on the effect of cooking on the shape, size and other features of starch grains that are visible with light microscopy, and these focus on only a few taxa, or on specialized cooking methods (e.g. Lamb and Loy, 2005; Samuel, 2000; del Pilar Babot, 2003). In order to better understand how the appearance of starches can be altered, we performed a series of experiments on Old World plant foods to document how different forms of cooking, such as fermenting, baking, boiling, grinding and parching, may cause changes to starch grains.

These experiments show that cooking damage to starch grains is recognizable, in comparison with undamaged starch grains or starch grains exposed to water. Though the starch grains are often too damaged to be identified to species, they are recognizable as cooked starch grains, thus providing a means for identifying cooking in the archaeological record. However, while some cooking methods produce unique changes to certain species, no methods produce identical results for each plant. Thus, for certain cooking methods to be accurately identified in the archaeological record, modern samples of the plant types of interest must first be subjected to cooking. In an effort to share this information, we have built a database of our starch images under all the different methods of cooking which is freely available at http://www.osresearch.net/~hollyf/starchdb/index.cgi.

2. Materials and methods

We chose 10 domesticated Old World plant species, mostly of Near Eastern origin, including four legumes and six grains from four Poaceae tribes (Table 1). Of the grains, we chose two of the top three most-produced grains (maize has the highest production rates, followed by wheat and rice), (USDA Annual World Production Survey, 2001–2002). We sought to include a variety of tribes and subfamilies in order to see if there are consistent patterns among these taxonomic groups. For each plant we examined the seed (caryopsis or pulse) as this is the plant organ with the highest starch content and the part most commonly used as food. The raw materials were purchased in bulk form at Whole Foods Market, and in the case of millet we confirmed the taxonomic identification by contacting their food manager, who contacted the farmer/supplier.

Prior to cooking, the native starches of the raw plants were examined and described in order to have a baseline against which to compare any possible changes due to cooking (Table 2). These descriptions were also compared against previously published reference collections (Piperno et al., 2004; Reichert, 1913, 1919; Seidemann, 1966). Each plant type was then subjected to four cooking methods – boiling, baking, parching and fermenting with extra variants in the time of cooking and whether the grain was ground or whole. In addition, the grains were popped – heated while dry until the pericarp split and the inner starchy endosperm was visible. All plant types were fermented with baker's yeast, and two grains and one legume were also ground and then fermented. All were ground and soaked in water at room temperature for 4 h to see if exposure to water without heat would change the starch grain morphology. See Table 3 for details on the cooking methods. After being cooked, a small part of each sample was then transferred to a microscope slide, either by scraping off a small portion with a scalpel and adding water to mount the slide (whole samples) or by transferring the liquid slurry by pipette (ground samples).

Each sample was examined under one of three microscopes. We used a Zeiss Axioskop 2 polarizing light microscope with attached AxioCam camera, linked to a PC running AxioVision software, a Leitz Laborlux 12 Pol S light microscope with attached Polaroid camera, linked to a PC running Polaroid DMC le software, or an Olympus CX41 with attached QImaging MicroPublisher 3.3 RTV camera, attached to a Mac G5 running QCapture software. At least five images of each slide were captured and the sample morphology was described in detail, with particular care taken to understand differences between the processed sample and the whole, uncooked form and from other cooking times and techniques. The degree of damage to the starch grain was likewise noted, with damage defined as swelling, 'lumpiness', loss of features like lamellae, fissures or dimples, and reduction or change in the extinction cross.

3. Results

Varying the cooking method produced some different changes among the analyzed starch grains. Starch grains exposed to water at room temperature (soaking, fermenting) generally changed very little, with only very slight swelling of the grain and a dimming or widening of the dark part of the extinction cross. Application of heat in the absence of external water caused quite extensive damage to the starch granules, usually extreme swelling and some cracking. Heating in the presence of water in general (boiling, baking) caused the starch grains to swell, lose distinctive features, lose their extinction crosses, and to appear softer. Longer cooking times universally caused more extreme swelling and loss of features until all of the starch grains were completely gelatinized and unrecognizable as starches. As they swelled, compound starch grains that previously had faceted margins grew rounder, whereas round, simple starch grains generally grew bumpy and wrinkled. Starch granules from ground samples showed more damage at shorter cooking times than those from whole samples, suggesting that the starches at the centers of whole caryopses or pulses were protected. Finally, the appearance of the extinction cross is often

Table 1
Plant species analyzed for their response to various cooking techniques.

Family	Subfamily	Tribe	Genus and species	Common name
Poaceae	Pooidae	Triticeae	*Triticum aestivum* L.	Hard red winter wheat
			Hordeum vulgare L.	Barley
			Avena sterilis L.	Oats
	Panicoideae	Paniceae	*Panicum miliaceum* L.	White proso millet
		Andropogoneae	*Sorghum bicolor* (L.) Moench	Sorghum
	Erhartoideae	Oryzeae	*Oryza sativa* L.	Rice
Fabaceae	Faboideae	Fabeae	*Lens culinaris* Medik.	Lentil
			Pisum sativum L.	Green pea
		Cicereae	*Cicer arietinum* L.	Chick pea, Garbanzo
		Phaseoleae	*Vigna radiata* (L.) R. Wilczek	Mung bean