

Starch grains on human teeth reveal early broad crop diet in northern Peru

Dolores R. Piperno^{a,b,1} and Tom D. Dillehay^{c,d}

^aArchaeobiology Program, Department of Anthropology, Smithsonian National Museum of Natural History, Washington, DC 20560; ^bSmithsonian Tropical Research Institute, Apartado Postal 0843-03092, Balboa, Republic of Panama; ^cDepartment of Anthropology, Vanderbilt University, Nashville, TN 37265; and ^dDepartamento de Antropología, Universidad Austral de Chile, Valdivia, Chile

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Previous research indicates that the Ñanchoc Valley in northern Peru was an important locus of early and middle Holocene human settlement, and that between 9200 and 5500 ¹⁴C yr B.P. the valley inhabitants adopted major crop plants such as squash (*Cucurbita moschata*), peanuts (*Arachis* sp.), and cotton (*Gossypium barbadense*). We report here an examination of starch grains preserved in the calculus of human teeth from these sites that provides direct evidence for the early consumption of cultivated squash and peanuts along with two other major food plants not previously detected. Starch from the seeds of *Phaseolus* and *Inga feuillei*, the flesh of *Cucurbita moschata* fruits, and the nuts of *Arachis* was routinely present on numerous teeth that date to between 8210 and 6970 ¹⁴C yr B.P. Early plant diets appear to have been diverse and stable through time and were rich in cultivated foods typical of later Andean agriculture. Our data provide early archaeological evidence for *Phaseolus* beans and *I. feuillei*, an important tree crop, and indicate that effective food production systems that contributed significant dietary inputs were present in the Ñanchoc region by 8000 ¹⁴C yr B.P. Starch grain studies of dental remains document plants and edible parts of them not normally preserved in archaeological records and can assume primary roles as direct indicators of ancient human diets and agriculture.

early diets | South America | food production

The Late Pleistocene to Middle Holocene Ñanchoc preceramic culture is known from 46 sites in the Ñanchoc Valley, a tributary of the Zaña Valley located at 500 m above sea level on the lower western slopes of the Andes (1–7) (Fig. 1). Recovered from hearths and floors in 13 excavated house structures dating from ~10,100 to 5500 ¹⁴C yr B.P. (~11,200 to 6000 calendar years ago or cal. B.P.) were the macrobotanical remains of a variety of plant cultivars along with gathered wild plants and the bones of various large and small animal species, providing evidence for a broad spectrum and mixed subsistence economy in a tropical dry forest setting (1–3, 6). Between 10,000 and 7000 ¹⁴C yr B.P. during the Late Paijan and Las Pircas phases, crop production was practiced close to small, permanent, circular houses. Associated with the plant remains during the subsequent Tierra Blanca phase between 7000 and 5000 ¹⁴C yr B.P. were stone hoes, larger grinding stones, plots for planting crops, small-scale irrigation canals and public mounds (site CA-09–04, Fig. 1), and rectangular houses (1–3, 6).

We examined 39 human teeth from Las Pircas occupations at four different sites [Table 1, supporting information (SI) Table S1]. The teeth were mostly isolated remains from adults and subadults that were securely embedded within intact and sealed house floors and directly associated with radiocarbon dates made on human bone, macrobotanical plant remains, and wood charcoal ranging from 8210 ± 180 to 6970 ± 60 ¹⁴C yr B.P. (Methods; Table 1; Tables S1 and S2; see ref. 1 for a more complete list of ¹⁴C dates from the sites). Three teeth were submitted to Beta Analytic Inc. for accelerator mass spectrometer (AMS) radiocarbon dating but insufficient protein remained for an age determination (Table 1). The isolated teeth

come from an estimated minimum of six to eight individuals, probably from both sexes; three complete individuals are also represented (Table 1).

We cannot state with much precision how much of an individual's lifetime the tooth calculus that was studied represents. Little information is available on the rate of calculus formation but because it accumulates over an individual's life if not removed, at least several years of diet is probably represented for each tooth studied. To remove dental calculus and examine and identify the starch in it we used nondestructive methods developed by ourselves and large modern reference collections housed in D.R.P.'s laboratory (Methods and SI; we also searched for phytoliths in the calculus preparations and found none, not surprising in view of the fact that the plants evidenced on the teeth and many others that were potentially available in the study region do not produce phytoliths in their edible parts).

Results

Most teeth yielded some starch grains and 13 contained them in significant quantities. Table 1 contains the kinds and number of grains retrieved from these specimens. Table S1 contains data on the teeth with fewer grains. Ubiquitous and consistently common over the 1,000-year period represented are four taxa, *Phaseolus*, *Arachis*, *Cucurbita moschata*, and *Inga feuillei*, indicating they were major dietary sources (Table 1) (Fig. 2 a–e). We cannot determine conclusively whether the *Phaseolus* starch is from a domesticated bean or whether it has a greater affinity to *P. lunatus* or *P. vulgaris* (Fig. 1 a and b for archaeological *Phaseolus*; Fig. S1 A and B for modern comparisons). It is generally considered that single domestications of the Andean lima and common bean occurred, respectively, in southern Ecuador/northern Peru and southern Peru/Bolivia (8–12). Three species of wild *Phaseolus* closely related to domesticated species, all of which were analyzed here, grow in Peru: *P. augusti*, *P. pachyrhizoides*, and wild *P. vulgaris*. The first two probably should be merged taxonomically and are thought to be the lima's wild ancestor (9, 12).

In most teeth samples, average and maximum *Phaseolus* grain size is considerably larger than in wild common bean, which can be excluded from representation on this basis (Table 2 and Table S3 for comparative modern data). However, there is a broad range of size overlap between wild and domesticated lima bean starch, and size values for all but one of the teeth fall within this overlap zone. In sample 19, dated to 6970 ¹⁴C yr B.P., average length and width of *Phaseolus* grains exceed those of all modern wild beans studied. Given the size similarities between wild and

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¹To whom correspondence should be addressed. E-mail: pipernod@si.edu.

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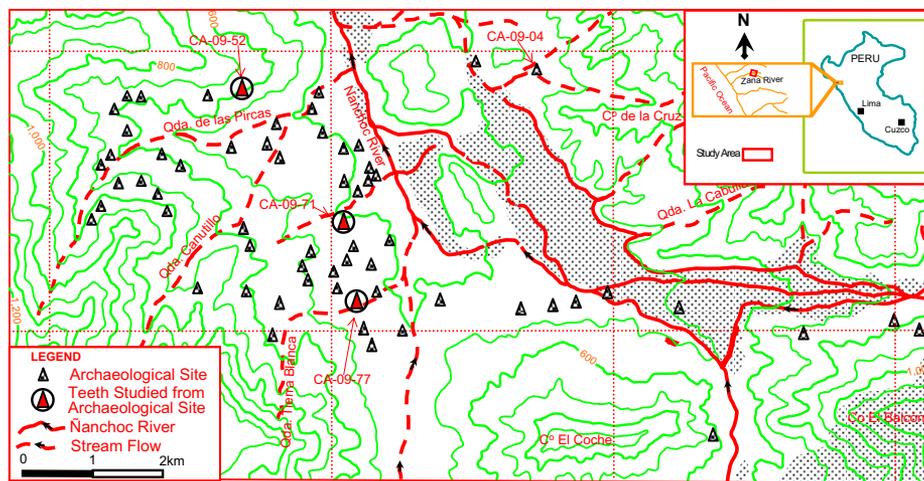


Fig. 1. Location of the Nanchoc Valley in northern Peru and archaeological sites discussed in this study.

domesticated lima bean starch, we prefer to exercise caution at this time with the interpretation of this single tooth showing a domesticated grain size. Starch from wild and domesticated *Phaseolus* species is too similar in morphology to attempt a species-specific identification on this basis (Methods and Fig. S1 A and B).

The ecology of wild *Phaseolus* does strongly suggest that the starch grains represent cultivars. Wild lima and common beans are found today in dry and thorny or humid forest at elevations between 1,800 and 3,000 m above sea level (asl) (9). The Nanchoc sites, located at 500 m asl in dry tropical forest, are considerably outside of the elevational range and native habitats of these plants. The previous earliest record for *Phaseolus* in South America was the domesticated *P. lunatus* from Chilca, southern coastal Peru, dated to 5600 ¹⁴C yr B.P. (ca. 6400 cal.

B.P.) (13). The earliest common bean remains, from Guitarrero Cave in central highland Peru (13), presently date to ca. 4600 ¹⁴C yr B.P. (ca. 5000 cal. B.P.). Considering this evidence along with the greater proximity of the Nanchoc region to the probable hearth of lima domestication, it may be more likely that the *Phaseolus* starch is from a lima rather than a common bean.

Previous work identified the early presence of a domesticated squash species, *Cucurbita moschata*, which likely is native to lowland northern South America, possibly Colombia. Distinctive, elliptical-shaped seeds dark brown in color that are unique to *C. moschata* were recovered from sites CA-09-77 and CA-09-27 where they were directly dated to 9240 ± 50 ¹⁴C yr B.P. and 7660 ± 40 ¹⁴C yr B.P., respectively (1). Starch grain data provide an independent line of evidence for this crop and broaden our understanding of how *Cucurbita* was consumed

Table 1. Types and Numbers of Starch Grains Recovered from the Nanchoc Teeth

Sample/ ¹⁴ C Age	<i>Phaseolus</i>		<i>Arachis</i>		<i>Cucurbita moschata</i>		<i>Inga</i> *	Legume	Globular	Bell	U #1, 2, 3, 4, 5, 6	Other	Total
	<i>n</i>	<i>n</i> (x̄ Size; range)	<i>n</i>	(x̄ Size; range)	<i>n</i>	(x̄ Size; range)							
Site CA-09-77													
2A, 6970 ± 60 HB	14	3 (8.7; 6-12)	11	(8.3; 6-12)	26	5	0	0	1	-----	11(1)	71 I	
29, 6970 ± 60 HB	55	2 (6; 5-7)	2	(7; 5-9)	4	13	5	0	5	-----	10(5)	91 M	
19, 6970 ± 60 HB	24	0	1	(12)	1	0	2	0	-----	7(4)	35 M		
68, #1, 7840 ± 40 PH+	93	0	4	(9.8; 8-11)	0	1	2	0	2	-----	12(2)	114 M	
68, #2, 7840 ± 40 PH	37	4 (7-7)	2	(13; 11-15)	2	13	9	1	-----	12(8)	80 M		
7, #2, 7840 ± 40 PH	3	5 (10.2; 7-14)	0	0	0	0	1	0	-2	----	2	13 M	
50, #1, 7840 ± 40 PH+	14	41 (11.9; 7-18)	0	2	0	0	See note	0	-----	41(8)	98 M		
50, #4, 7840 ± 40 PH	22	6 (9; 6-13)	2	(6-6)	14	1	12	1	15 2 2 - -	-----	13(3)	90 M	
56, #4 7840 ± 40 PH+	5	2 (13; 8-17)	0	0	0	0	0	0	-----	11(9)	18 M		
**59, 7840 ± 40 PH	23	3 (13.3; 12-15)	0	0	4	0	0	0	1 2 - 1 - 1	-----	4	42 M	
58, #1, 7840 ± 40 PH	22	1 (12)	0	0	0	0	0	0	- 2 - 2 2 -	-----	4	27 M	
58, #3 7840 ± 40 PH	3	3 (8.7; 8-9)	4	(8.5; 6-11)	14	0	7	1	- 2 - 2 2 -	-----	17	55 M	
Site CA-09-28													
1A 8210 ± 180 WC	4	1 (9)	9	(9.8; 7-12)	1	0	0	0	-----	-----	9	15M	

Note: ¹⁴C dates from CA-09-77 are derived from materials (HB = human bone, PH = charred peanut hull) directly associated with teeth from hearths sealed in individual intact floors (ca. 3 cm thick) of buried house structures (see Table S2 and refs. 1, 6 for more details). For CA-09-28, the associated date is on wood charcoal. +Teeth were submitted to Beta Analytic Inc. for AMS radiocarbon dating but insufficient protein remained for an age determination. *Identified as *Inga feuillei*. The category "Legume" represents oval grains that occur in *Phaseolus* and also in other Fabaceae genera. "Globular" grains are consistent with those in *Arachis* and *Inga* and also occur in a variety of non-legume taxa. **Three grains typical of underground organs occurred. In sample 50, #1 many of the "Other" grains occurred in clumps and were difficult to describe in detail but they were globular in shape as in peanut and/or *pacay*. "Bell" grains that occurred in small numbers on some teeth are consistent with manioc but a positive identification is not possible. Numbers in parentheses in the "Other" category represent gelatinized grains that were too damaged from heat to identify. Letters in bold after Total *n* indicate type of tooth (I = incisor, M = molar). Teeth 50 and 58 are from sub-adults. Others are indeterminate as to the approximate age of the individual. Tooth 1A is from a complete skeleton.

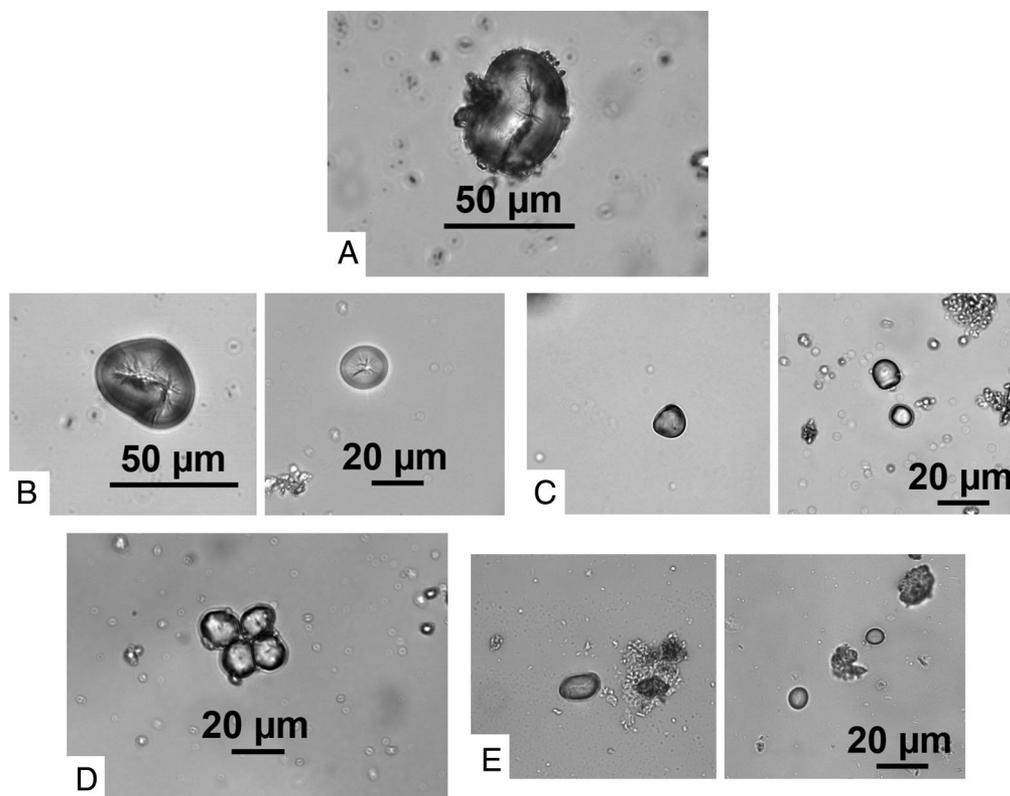


Fig. 2. (a) An oval starch grain from *Phaseolus* with a longitudinal fissure and lamellae from tooth 29. (b) *Left*, an oval starch grain from *Phaseolus* with a ragged longitudinal fissure and lamellae. *Right*, a round starch grain from *Phaseolus* with a stellated fissure and lamellae concentrated near the edge. Both grains are from tooth 50, no. 4. (c) Bell-shaped grains from teeth 1A (*Left*) and 2A (*Right*) with "pleats" (unique kinds of lamellae) and pressure facets from *Cucurbita moschata* fruit flesh. (d) A cluster of four starch grains from peanut nuts from tooth 50, no. 1. The grains are globular in shape and have projections/knobs at the edge, slight fissures, and tiny starch grains attached at the periphery. (e) Oval (*Left*) and elliptical (*Right*) starch grains with white longitudinal fissures from *Inga feuillei* from samples 50, no. 4 and 58, no. 3.

during early periods. Many teeth contained distinctive bell-shaped grains diagnostic of the flesh of *C. moschata* fruits (Fig. 2c; Fig. S2a) (seeds from the genus have no starch). In morphology and size the grains are identical to those found in the flesh of traditional modern fruits of *C. moschata* and they are unlike those of other *Cucurbita* species, including *C. sororia*, a wild taxon closely related to *C. moschata* (14) (Methods; SI). *Cucurbita* flesh produces significantly less starch than legume seeds, thus it is likely to be underrepresented in the records. The evidence firmly indicates that at Nanchoc early domesticated *Cucurbita* was routinely consumed and not used primarily as a utilitarian or industrial plant (e.g., as containers and fishnet floats). Moreover, as all wild *Cucurbita* species have extremely bitter, nonedible flesh, human selection for the major domestication gene that codes for nonbitter fruits by 8000 ¹⁴C yr B.P. is implied.

Another plant evidenced through previous research is the peanut. Hulls from *Arachis* sp. recovered from CA-09-77 were directly dated to 7840 ± 40 ¹⁴C yr B.P. (1). The hulls were not exact morphological matches for modern *A. hypogaea* but neither did they conform to a known wild species. They almost certainly represent cultivars because they occur far outside the natural distribution of the genus in southern South America (15). Peanut nuts were not preserved in the macrobotanical records; however, starch grains diagnostic of *Arachis* nuts (hulls contain no starch) are present on many teeth (Table 1; Fig. 2d; Fig. S2b). In their morphological and size characteristics they are matches for modern *Arachis hypogaea* (compare peanut data in Table 1 with that for modern varieties in Table S3). We also studied two peanut hulls retrieved from sites CA-09-77 and CA-09-50 that

date to ~8000 to 7800 ¹⁴C yr B.P. and 7000 ¹⁴C yr B.P., respectively, to ascertain whether any starch grains from decayed nuts survived in them. These specimens also yielded grains like those in *A. hypogaea*.

We examined a diploid wild species, *A. williamsi*, which is closely related to *A. hypogaea*. It has starch grains with a mean length of 6.6 μm and a maximum length of between 4 and 10 μm. This length is much smaller than grain size on the teeth and in different varieties of modern *A. hypogaea*, where maximum length was ≥18 μm and average length was from 9.5 to 10.1 μm (Table S3). Grains from *A. williamsi* also lack surface decorations found in *A. hypogaea*. We also studied three wild *Arachis* species, *A. burchelli*, *A. marginata*, and *A. retusa*, which are from a different taxonomic section of the genus that is more distantly related to *A. hypogaea*. They all exhibited markedly different starch morphological attributes (bell shaped instead of globular) when compared with *A. hypogaea*, *A. williamsi*, and the archaeological grains, providing more evidence that the Nanchoc peanuts were closely related to *A. hypogaea*. This may be a case where different traits indicative of domestication, such as hull morphology and nut starch grain qualities in these peanut examples, were not transformed from their wild states simultaneously or at the same rate by early human selection pressure, resulting in early cultivars that have a mixture of wild- and domesticated-type features. Analysis of the direct wild progenitors of the peanut, thought to be *A. ipaensis* and *A. duranensis* or *A. helodes* (15, 16), will shed more light on this issue.

Inga feuillei (*pacay*) is an important tree crop that was well used on the Peruvian coast by 4500 ¹⁴C yr B.P. (17–19). It is probably native to the lowland eastern slopes of the Andes and

Table 2. *Phaseolus* starch grain morphology and size on the Ñanchoc teeth

Site and specimen	Age	Oval, no damage			Oval damaged			Round with lamellae		Round with fissures	
		Length, μm	Width, μm	<i>n</i>	Length, μm	Width, μm	<i>n</i>	Diameter, μm	<i>n</i>	Diameter, μm	<i>n</i>
CA-09-77											
19	6970 \pm 60	39 (32–53)	29 (23–39)	5	37 (28–52)	30 (20–51)	15	30 (29–30)	2	14 (11–17)	2
29, #1	6970 \pm 60	29 (18–50)	23 (9–36)	5	27 (19–47)	21 (8–31)	19	24 (20–29)	4	19 (15–23)	2
27, #2	6970 \pm 60	44	38	1			0		0		0
10, #1	6970 \pm 60				20 (19–21)	16 (14–17)	2		0		0
2A	6970 \pm 60	35 (26–41)	25 (21–32)	7	24 (19–31)	17 (15–20)	4		0	23 (13–32)	3
29, #2	6970 \pm 60			0	29 (26–32)		2		0		0
29, #3	6970 \pm 60			0	34	27	1		0		0
68, #1	7840 \pm 40	29 (19–44)	25 (17–37)	13	29 (16–41)	24 (13–36)	17	27 (20–36)	7	15	1
68, #2	7840 \pm 40	29 (23–39)	23 (17–33)	9	29 (16–46)	24 (12–42)	18	21 (15–26)	9	22	1
7, #1	7840 \pm 40			0	39 (36–43)	26 (24–27)	3				
7, #2	7840 \pm 40	27	24	1	28	22	1		0	26	1
50, #1	7840 \pm 40	33 (27–39)	23 (22–26)	3	23 (18–27)	21 (16–24)	7	41	1	14 (10–16)	3
50, #4	7840 \pm 40	34 (21–41)	25 (15–32)	4	28 (18–42)	21 (13–29)	12	28 (26–29)	2	15 (12–19)	4
50, #2	7840 \pm 40	20		1	22	18	2	26	1	18 (16–21)	3
56, #4	7840 \pm 40			0	36 (30–45)	27 (24–31)	4	31	1		0
59	7840 \pm 40			0	32 (22–52)	24 (17–44)	20	47	1		0
58	7840 \pm 40				29 (17–48)	24 (12–41)	19		0		0
58, #1	7840 \pm 40			0	29 (17–48)	24 (12–41)	19	16 (14–17)	2	37	1
58, #3	7840 \pm 40	28 (27–29)	23 (22–23)	2			0	37	1		0
CA-09-52											
117	8080 \pm 70			0			0	19	1		0
113	7920 \pm 120				27	16 (13–19)	2				0
15	7920 \pm 120	46	32	1	31 (24–41)	24 (14–31)	5	36	1	22	1
3A	7920 \pm 120	24	18	1	34	29	1		0		0
CA-09-28											
1A	8210 \pm 180	38	22	1	29 (22–36)	21 (20–21)	2	33	1	10	1

Phaseolus oval grains in the “no damage” category exhibit no signs of damage from heat or other sources. Oval grains in the “damaged” category exhibit heat damage such as altered or no extinction crosses and damaged fissures (Figs. S3 a–d and S4 a–d). “Round with lamellae” grains are those without fissures and with lamellae covering the entire grain. “Round with fissures” are the smaller round grains that have a variety of fissure types, including stellate, and are without lamellae or have lamellae concentrated in a thin, dense band at the periphery of the grain. The presence and proportions of these different grain types on the teeth were also matches for the genus *Phaseolus* and no other legume taxon. In the size categories, the first number is the mean and the numbers in parentheses are size ranges for the grains.

it can be grown at much higher elevations, which made it important to the Inca (20). *Pacay* has large edible pods with a sweet, white pulp. Starch identified from this plant derives from the seeds; the pulp does not contain starch (Fig. 2e; Fig. S2c). This is clear evidence for early tree crop exploitation in the Ñanchoc region.

A variety of other starch grains occurred in lower quantities and just a few of them appear to be from underground organs (Table 1). Three grains were consistent with those from manioc roots although a positive identification is not possible from the grain types present (21). Macrobotanical remains of manioc were retrieved from the sites (6). Low overall frequencies of starch from underground organs suggest they were infrequently consumed. Other grains that occurred in low numbers, including six types of unknown but distinctive-looking starch (U nos. 1–6), are conceivably from foods such as cacti and tree fruits that are not represented in our modern collections. It also should be pointed out that some potentially important food sources that were well used during the later pre-Columbian period in the study region do not produce starch or have nondiagnostic grains, thus their dietary contribution cannot be studied. They include domesticated tree fruits of various *Annona* species (guanabana, custard apple) and guava (*Psidium guajava*), other fruits (*Bunchosia armeniaca* and *hartwegiana*; *Solanum quitoense* or *naranjilla*), and the lupine bean (*Lupinus mutabilis*).

Discussion

Starch grain data from dental remains can inform a number of important issues concerning early human diets and the transition from hunting and gathering to agriculture. It has long been discussed, for example, whether the earliest plants taken under cultivation in some areas of the world were dietary items or used for nonfood purposes. In the Americas, at least four different species of *Cucurbita* native to North America, Mesoamerica, and South America are now known to have been among the first crop plants in these regions, and it has recently been proposed that early cultivation and diffusion of *Cucurbita pepo* ssp. *ovifera* in the eastern United States were motivated much more by the utility of the plant for its nonfood uses than by its role in the diet (22, cf. ref. 23). In contrast, the Ñanchoc evidence clearly shows that *C. moschata* was commonly eaten by some of the first farmers in Peru.

Overall the evidence indicates that by ca. 8000 ¹⁴C yr B.P. (ca. 8600 cal. B.P.) an effective farming system employing a range of seed, vegetable, tree, and root crops provided balanced, nutritious, and stable diets to the inhabitants of the tropical western slopes of the Peruvian Andes. The ubiquity of plants at Ñanchoc that would come to typify later agricultural systems suggests that regional dietary patterns began to emerge not long after food production and crop dispersals began. The multifaceted botanical, settlement (sedentary and seminucleated communities),

landscape (irrigation canals and furrowed fields), and artifactual data (e.g., stone hoes, grinding stones, storage pits) (1–7) show that Ñanchoc societies cannot be characterized as casual food producers who were deriving only negligible dietary inputs from cultivated plants. The evidence from here and some other well-studied regions in the Neotropics (17, 24, 25, 26) instead indicates an early Holocene development of food production together with an emergence not long afterward—at ~8000 to 7000 ^{14}C yr B.P.—of subsistence economies that included significant amounts of cultivated and domesticated products. Thus, although large sedentary and nucleated villages do not occur in the Americas until after 5000 ^{14}C yr B.P., earlier food production in some areas was practiced by people who, while still integrating planting with foraging, were committed farmers who had taken the first steps along the pathway to full-fledged agriculture.

A routine cultivation (a persistent cycle of sowing and harvesting) of plants that provide stable and significant dietary inputs, but that do not acquire all or even some of the phenotypic traits typical of the domestication syndrome for hundreds to thousands of years (known as predomestication cultivation), is a pattern increasingly being documented at the beginning of agriculture in the Old World (27, 28). The issue has received less attention thus far in the New World, in part because early macrobotanical remains from the tropics are rare, but some potential examples exist from highland Mexico (25). The peanut evidence from Ñanchoc points to an instance of predomestication cultivation (hulls lack features of modern *A. hypogaea*) possibly together with a time-transgressive appearance of different domesticated traits in this plant (starch grains are matches for modern *A. hypogaea* and not wild *Arachis* studied), suggesting that domestication in some major New World crops may also have been a protracted and complex process. Possible instances of predomestication cultivation should be sought more widely in the New World using multifaceted archaeobotanical data sets and allied paleoecological evidence when possible. Much more evidence is needed but it is possible that as in the Old World, traditional, domestication-based concepts of agricultural origins—those that posit that a suite of fully domesticated plants formed the basis for the first effective food production systems and initial spread of crops—may have to be adjusted.

In conclusion, the Ñanchoc evidence elucidates in a number of ways early agricultural lifeways in the Andean region. While, on the present evidence, most of the Central Andes between 10,000 and 8000 ^{14}C yr B.P. were occupied by a wide variety of maritime and terrestrial foragers that were still organized in relatively small, mobile groups, some populations such as those in the Ñanchoc Valley were adopting more sedentary life styles and becoming farmers. The incorporation into subsistence economies of domesticated plants and other cultigens during this period did not immediately lead to a full-scale agricultural economy, as Ñanchoc people continued to hunt, fish, and gather wild plants. Managing a wide range of food sources across several closely juxtaposed ecological zones may have helped to reduce subsistence risk. By ~6500 to 6000 ^{14}C years ago, the emergence of a planned aggregated community and small-scale public monuments in the area are evident, anchored by more intensive food production systems that now included irrigation canals and crop field systems (2, 5). These developments, in turn, created new ways of life, new perceptions of public and private spaces, and new relationships between members of a community, which represent some of the initial pulses toward the rise of Andean civilization (5). More complex ideological, technological, and subsistence advances that took place after 5500 ^{14}C yr B.P. in Peru (18–19), including a shift to agricultural dependence, are probably direct extensions of the Ñanchoc developments.

Methods

Starch Grain Sampling of Teeth. Our sampling strategy used methods non-destructive to the teeth and the starch (29). First, teeth were brushed with a soft tooth brush and water to remove adherent soil and other particles. A dental pick was used to scrape areas of teeth with visible calculus, and the residue was transferred directly to a microscope slide where a few drops of water had been placed. On many teeth this procedure was done from three to five separate times by sampling different areas of the specimen such as the crowns of molars and the gum line and making new microscope mounts each time. The plaque could at times be seen to be yellowish in color when it was transferred to the slide. Sampling was stopped when no additional starch grains were recovered. Before the coverslip was put on, one drop of 50% water/glycerine was added to the residue-water suspension to retard drying and allow grains to be more easily rotated when encountered. This light solution of glycerine does not alter starch morphology. The non-use of chemicals ensures that starch grains will not suffer damage during and after their removal from dental remains.

Archaeological peanut hulls were examined by adding a few drops of water to the inside of the specimens using a pasteur pipette, gently washing the hull wall with the water, and transferring the liquid to a microscope slide.

Starch Grain Identification. We used a large modern reference collection numbering ~500 economic and other plant species from 50 different families along with published sources (21, 24, 30–35). Our modern collection includes all major and many now-minor crops and other plants known or thought to have been used in Latin America/the Andean region during prehistory and many wild species closely related to crop plants such as: (1) the wild progenitors of lima and common beans from South America and Mexico; (2) other wild Fabaceae including *Inga*, *Canavalia*, *Vigna*, and *Macropitilium*; (3) wild *Arachis*; and (4) wild *Cucurbita* native to southern Central America and South America (*C. maxima* ssp. *andreaana*, which is the wild ancestor of *C. maxima* and *C. argyrosperma* ssp. *sororia*) (see also ref. 21 for further details on the collection). *C. sororia* is closely related to the as yet undiscovered wild ancestor of *C. moschata* (14).

We studied a diverse collection of modern, traditional lima and common bean land races from Peru, Mexico, and other areas of Latin America. Our results indicate the following: (1) there is considerable overlap in grain size of wild lima beans, domesticated lima beans, and domesticated *P. vulgaris*; (2) grain size in wild common beans appears to be significantly smaller than in both domesticated common and lima bean, and (3) there is considerable morphological similarity among wild and domesticated species in *Phaseolus*, making species-specific identification difficult on this basis (Table S3; Fig. S1 A and B).

Our starch keys and classification system emphasize shape, surface, and size attributes known to be useful in identification (21, 24, 30–35) (see also Figs. S1–S5). To confirm identifications of *Phaseolus*, *Arachis*, *C. moschata*, and *I. feuillei* we took advantage of the fact that each, along with genera and species closely related to them, produce more than one type of starch grain, resulting in taxon-unique starch population signatures when a multiple grain analysis is undertaken. Archaeological sample sizes from individual teeth often permitted such an analysis, and these population signatures in addition to the occurrence of diagnostic, individual grain types robustly indicate the presence of these four major taxa (Figs. S1–S5).

We encountered partially and completely gelatinized grains, presumably a result of cooking, on many of the teeth. Some were identifiable because they retained their shape and surface characteristics, and some still exhibited an extinction cross. Recognition of these grains was based on cooking experiments we carried out (below) and other research (34–36).

Cooking Experiments. We cooked whole common and lima beans by placing them in a test tube and then into a hot water bath for times ranging from 15 to 45 min. After 15 min many grains expanded to three times their normal size, lost their lamellae and fissures, and exhibited no or damaged extinction crosses. However, many retained their characteristic shape, size, and surface attributes and their extinction crosses, showing no signs of damage (Fig. S3 a and b). Additionally, a small proportion of damaged grains still exhibited the characteristic oval to kidney *Phaseolus* shape and had lamellae and remnant fissures. The majority of starch grains in samples that were cooked for 45 min lost lamellae and fissures, but many retained the characteristic *Phaseolus* shapes, and lamellae and remnant fissures were still visible on a few (Fig. S3c). Most still polarized, exhibiting the diffuse extinction cross arms that are characteristic of heat damage (Fig. S3d). Cooked grains that retain *Phaseolus* shapes and have lamellae or remnant fissures are recognizable as *Phaseolus*, and they routinely occurred on the teeth (Fig. S4 a–d).

We made comparisons of starch grain size in uncooked and cooked beans, in the latter measuring only grains that were recognizable as *Phaseolus*

(Table S3). Cooked grains were either marginally larger or smaller than uncooked specimens. Our analysis of roasted and unroasted peanuts indicates that starch from roasted specimens does not suffer morphological damage or size alteration, presumably because the hull offers a good deal of protection. We expect we will achieve a better understanding of these issues when experiments are carried out using cooking methods more analogous to prehistoric techniques, such as pit roasting and hot rock boiling. Nonetheless, it is clear that starch grains from cooked foods are surviving in an identifiable form on ancient human teeth. They have also been recovered from blackened food residues in Early Formative ceramic cooking vessels from Ecuador and stone tools from Ecuador and New Guinea (34–36).

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