tive, is how best to elucidate microbial function—that is, how they make their living. No one technique is yet sufficiently well developed to do this; however, functional gene microarray systems, which look for expression of genes coding for enzymes involved in specific functions, are currently under development (*13*).

An associated challenge is to determine the role of community microbial activity in shaping geochemical consequences. Geochemical interest in microbial activity is often restricted to a single metabolic function with a relevant geochemical consequence, such as metal sequestration (*5, 7*). However, the microbe responsible for that geochemical impact likely exists within a microbial community or consortium (*12*). The by-products from one organism's metabolic pathway are the nutrients of the next strain. Both the series of metabolic reactants and products associated with a microbial community, and the reaction energetics involved, are therefore likely to differ from one location to the next.

Thus, even though commonality of metabolic pathways ensures widespread occurrence of certain microbially driven geochemical processes in different environments, variability in microbial consortia and microgeochemical conditions will selectively refine that geochemical impact. In doing so, they may create community-specific microbial fingerprints on geochemical processes.

Microbial growth and activity can only proceed through inputs of energy, and are thus constrained geochemically to reactions that are thermodynamically feasible. High-resolution analytical tools for geochemistry and culture-independent molecular microbial techniques have yielded exciting insights. Today, microbial activity is viewed to play an important—and quantifiable—role in aqueous geochemistry. New molecular techniques for evaluating microbial functional activity will provide key information on how microbes engineer geochemical processes, and how they, in turn, are constrained by the geochemical world in which they find themselves.

Systematic examinations of the links between genome and geochemistry will explore gene expression (that is, function) and determine reaction kinetics of microbial communities growing under differing geochemical and physical conditions (*14*). Such studies will provide microbial fingerprints for important geochemical processes under microbial control. Moreover, such studies should help to quantify the microbial influence on important aqueous geochemical processes, determine the linked controls for these key processes, and show how feedback between microbial ecology and geochemical conditions influences the geochemical outcomes.

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ARCHAEOLOGY

Invisible Clues to New World Plant Domestication

Vaughn M. Bryant

Decades ago, excavations in the Tehuacan Valley of Mexico (*1*) convinced many archaeologists that they now had physical proof of how, when, and where plants were first domesticated in the New World. The proof came in the form of preserved seeds and fruits, maize kernels and cobs, fibers, and the rinds of cultigens found in cave soils and in preserved human feces originally dated as early as 7500 to 9000 years old. Little did these archaeologists realize that the puzzle of New World plant domestication was far from solved. Decades later, they would learn that the most critical clues come not from the large and visual remains of plants, but from tiny microscopic particles that most archaeologists unknowingly discarded.

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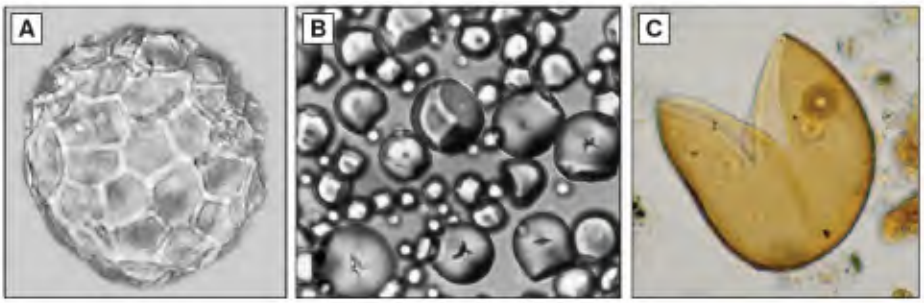
Fortunately, a few researchers (*2, 3*) were not convinced by the traditional story of New World cultigen origins. Piperno and a few others devoted more than three decades to searching the archaeological soils of Central and South America for microscopic phytoliths (plant crystals), tiny starch grains from domesticated plants, and fossil pollen (see the figure). As noted by Piperno and Stothert on page 1054 of this issue (*4*) and by Piperno and Pearsall in a recent book (*5*), these microscopic traces of plants reliably record the earliest use of domesticated plants.

Early speculation about the origins of New World plant domestication focused on the upland regions of Mexico and South America. These regions were favored because they were easy to reach, often contained caves or rock shelters filled with preserved plant remains, and had yielded previous successes that ensured



Archaeology under the microscope. (**A**) Radiocarbon-dated 10,000-year-old phytolith (diameter 100 μm) of domesticated *Cucurbita*, collected from soil at the Vegas Site 80 in Ecuador. (**B**) Reserve starch grains from the root of a modern manioc plant. (**C**) Maize pollen grain (diameter 75 μm) from cultural levels of the Kob Site, Belize, radiocarbon dated to ~5000 years ago.

CREDIT: PANELS A AND B, D. PIPERNO; PANEL C, J. JONES

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continued funding. Few archaeologists were willing to search for the origin of plant cultigens in lowland and jungle regions, where seeds, wood, rinds, and cobs did not preserve well. Many also wondered how tropical lowlands could have supported foragers making the switch to early sedentary farming. And most believed incorrectly that all lowland and jungle soils were infertile, similar to ones known from the non–flood plain regions of the Amazon. But Piperno and a few others remained convinced that the long search for cultigen origins had focused on the wrong areas and the wrong kind of clues.

It is true that, except for charcoal, the visual evidence of plant remains quickly disappears in most tropical soils. But some clues remain, if you know where to look. By the early 1990s, Piperno (*2, 6*) had shown that plant phytoliths were plentiful in the lowland soils of many regions of Central and South America. She noted that the size and shape of phytoliths were often unique to a family, genus, or species of plant (see panel A of the figure). Piperno and Pearsall then led the way in developing phytolith keys to a wide variety of New World cultigens and tropical plants (*5*). Armed with this knowledge, scientists began to search for microscopic clues in the soils of early, well-dated archaeological sites throughout Central and South America.

New studies based on phytolith data (*3, 6*) soon began to contradict the long-held theory that plant domestication began in upland regions, where many envisioned cave-living foragers switching to raising cultigens (*1*). Archaeologists challenging the new discoveries pointed to potential errors in dating, saying that the phytolith evidence could not possibly be as old as the dates indicated. To quell the critics, phytolith researchers developed new ways of dating tiny bits of carbon trapped inside phytoliths as they were formed.

The new techniques, which use accelerator mass spectrometry (AMS) dating, require the careful collection and separation of many phytoliths (*4*). Precise phytolith identification is also critical. Piperno and Stothert (*4*) collected and measured phytoliths from more than 150 mature fruits from wild and domesticated species of *Cucurbita* (squash and gourds) grown in 100 different locations. The phytoliths from domestic species were substantially larger than those from wild species. The authors then used phytolith size to confirm that domesticated *Cucurbita* were grown and used during the early Holocene in coastal Ecuador, between 9000 and 10,000 years ago.

Piperno (*7*) has also been at the forefront of searching for archaeological evidence of cultigen starch grains. In tropical regions of Central and South America, root crops—including yams, sweet potatoes, and manioc—are the mainstay of many indigenous cultures. These high-calorie tuber crops grow well in the wet soils and areas created by recently cleared tropical forests. But it has been difficult to prove when and where these plants were first domesticated. Tuber-producing crops do not carbonize well; furthermore, most produce small amounts of pollen and do not produce diagnostic phytoliths. However, they do produce copious numbers of water-insoluble granules called "reserve starch grains" (panel B), which preserve well on the surfaces of food preparation implements and in many types of tropical soils. Starch grains from tuber cultigens have recently been identified on early Holocene grinding stones used in Colombia and central Panama (*8*).

In 1957, the British pollen analyst Dimbleby (*9*) stated that soils with a pH above 6 are virtually useless for fossil pollen studies. Because the pH values of almost all tropical soils exceed 6, pollen analysts have for decades rarely searched the tropical soils of Central or South America. I was one of those pollen analysts who spent more than 30 years working at sites in Mexico and South America. I never found well-preserved pollen in any of those sites.

To the astonishment of many, Jones and colleagues recently recovered pollen from key archaeological sites in Central America (*10*). His pollen data confirm the use of early cultigens, including maize and manioc, from the San Andres site in the Mexican tropical lowlands near La Venta, Tabasco. Radiocarbon dating shows that the archaeological deposits containing cultigen pollen are 5800 to 6200 years old.

Perhaps DNA studies of soils in archaeological sites may soon replace our current techniques. Until then, our best New World records for cultigen origins are coming from the invisible clues: phytoliths, starch grains, and fossil pollen.

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IMMUNOLOGY

Regulating the Regulators

Fiona Powrie and Kevin J. Maloy

Recently, there has been an explosion of interest among immunologists in a subset of T lymphocytes that prevent harmful immune pathology. These regulatory T cells (T_R), most of which express the activation marker CD25, constitute a small number (5 to 10%) of the total population of $CD4^+$ T lymphocytes present in healthy individuals. $CD4^+$ T_R cells prevent a number of immune-mediated diseases, including autoimmune disorders, transplant rejection, and inflammatory bowel disease (*1*). Recent studies including two papers in this issue, by Hori *et al.* (*2*) on page 1057 and Pasare and Medzhitov (*3*) on page 1033, are beginning to elucidate T_R cell biology in more detail, particularly aspects of their differentiation and functional capabilities. These studies emphasize that T_R lymphocytes do not act in isolation, but are themselves influenced by cells of the innate immune system. An equilibrium is thereby established that allows effective responses against dangerous microbes while minimizing immune pathology.

The identification of transcription factors that direct the differentiation of naïve $CD4^+$ T cells into functionally distinct T helper 1 (T_H1) and T_H2 cells has transformed our understanding of the molecular basis of $CD4^+$ effector T cell responses (*4*). The study by Hori *et al.* (*2*) identifies the forkhead/winged helix transcription factor Foxp3 as a master regulator that promotes T_R cell differentiation. These investigators noted that both humans and

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