COPROLITES AND ARCHAEOLOGY: THE MISSING LINKS IN UNDERSTANDING HUMAN HEALTH

VAUGHN M. BRYANT¹ AND KARL J. REINHARD²

¹ Department of Anthropology, Texas A & M University (TAMU 4352), College Station, TX 77843-4352, e-mail, vbryant@ tamu.edu;
² School of Natural Resources, University of Nebraska, Lincoln, NE 68588, e-mail, kreinhard1@mac.com

Abstract—The study of human coprolites (feces) is just over 100 years old. During the last half of that period these studies have greatly advanced our understanding of the lives, economy, and health of our past ancestors. Originally, the focus was on the identification and significance of recovered bone fragments and plant macrofossils in coprolites. That later expanded into searches for pollen, phytoliths, hairs, feathers, endoparasites, starch, and other types of micro debris that one could recover in coprolites. Most recently, advances in molecular biology have enabled searches for steroids, blood typing, DNA, and microbes present in coprolites and the implications each type of evidence can offer about the individuals who produced them. Currently, the analysis of groups of coprolites from specific regions of the American Southwest reveals ancient diets that were high in fiber, rich in food diversity, and relied on calories from plants containing inulin, not starch. Those data are now being used to gain a better understanding of why certain Native American ethnic groups suffer high rates of obesity and type II diabetes when eating traditional Western diets rather than the types of ancient diets eaten by their ancestors.

INTRODUCTION

Coprolite analysis (the study of ancient feces) is now providing new insights into the diets and health of ancient cultures, and it is revealing the blood types, sex, and genetic haplotypes of the people who created those ancient fecal deposits (Sutton et al., 2010) The recovery and analysis of mtDNA, steroids, and occult blood in human coprolites offer exciting new potential for studies of diet preferences based on gender and ethnicity as well as the ancient world's distribution of peoples with different blood groups (Poinar et al. 2001; Sobolik, 2003). In spite of its recognized importance today, the examination of coprolites from archaeological sites is a relatively new area of investigation, being just over a century old.

HISTORY OF ARCHAEOLOGICAL STUDY OF COPROLITES

From the beginning of studies in archaeology, researchers had to rely on crude and often inaccurate guesses as to the health, cultures, and especially the diets of ancient peoples. Occasionally, human coprolites would be recovered at some site, but often they were either not recognized or discarded because they were considered unsanitary and certainly not the type of object one would put on display in a museum. So, these invaluable capsules of human waste containing precise information about the people who left them behind and the health and diets of those individuals were mostly ignored or discarded (Bryant, 1974). Some of the early researchers tried to examine a few human coprolites using a variety of methods, including soaking them in water or mild detergents, pulling them apart, or carefully grinding them through various sizes of screens. Nevertheless, what was needed during this early period was a new approach to the study of coprolites and, more importantly, a greater recognition of the importance of this new technique among the scientific community. Both of those aspects did not occur for more than 60 years after the first suggestion that coprolites were a worthy field of study.

HISTORY

The first historical record of coprolite studies might be credited to J. W. Harshberger (1896), a botanist at The University of Pennsylvania who suggested that the undigested seeds and bones found in the feces of prehistoric humans might offer a clue to the types of foods eaten. Just over a decade later, Young (1910) was the first to examine human coprolites collected from areas in Salts and Mammoth caves in Kentucky. By pulling the dried coprolites apart he found they contained sunflower

seeds and hickory nut shells and concluded that those items may have been important diet components. In that same year, Jones (1910) analyzed fecal material recovered from the intestines of mummies in Nubia and found they contained grape and melon seeds, barley husks, and various types of plant fibers. Soon after, Netolitsky (1911a, b, 1912) identified rodent and fish bones, sedge tuber remains, and grass seeds in coprolites from Egyptian mummies. Warren (1911) examined the visceral area of a buried Bronze Age skeleton in England and found it included blackberry, rose, and saltbush seeds. Although not officially coprolites, Warren's study was the first to demonstrate that the contents of fecal remains, and by inference diets, could still be recovered and examined from burials. Loud and Harrington (1929) were the next to report on the analysis of human coprolites. From their study of coprolite remains in Lovelock Cave, Nevada, they suggested that prehistoric diet patterns included a variety of wild seeds and plant fibers.

During the next three decades of this initial, coprolite experimental phase, Volney Jones (1936) looked at coprolite material recovered from the Newt Kash Hollow Shelter in Kentucky and found it contained many of the types of plant remains found in the shelter's deposits. Thus, the plant remains from both the site and coprolites, consisting of seeds from goosefoot or lambsquarters (*Chenopodium berlandieri*), erect knotweed (*Polygonum erectum*), maygrass (*Phalaris caroliniana*), sumpweed or marsh elder (*Iva annua*), and sunflower (*Helianthus annuus*), supported his conclusions that the ancient shelter dwellers had been part of the Eastern Agricultural Complex that focused on both gathering wild foods, and planting the seeds of the native plants. During this same period, Wakefield and Dellinger (1936) examined dried fecal remains found in a mummy recovered from a rockshelter site in Arkansas and found they contained the food remains of sumac seeds and acorns.

This early half-century period of initial coprolite studies ended with a few additional studies during the 1950s with the work of Sperry and Fonner, who found mesquite (*Prosopis*) and burro weed (*Isocoma*) seeds, antelope hair and bird feathers in prehistoric human coprolites from Danger Cave (Jennings, 1957). Webb and Baby (1957) looked at a few human coprolites from caves in eastern Kentucky and found results similar to the earlier work of Jones, which included evidence the early cave inhabitants probably ate diets of sunflower and goosefoot seeds as well as various types of insects. In his initial excavations at cave sites in Tamaulipas, Mexico, Richard MacNeish (1958) found ancient human coprolites containing food items such as maguey fibers (*Agave*), squash seeds (*Cucurbita*), insect fragments and pieces of snail shells.

The study of human coprolites entered a new phase beginning in the mid-1950s with the initial work of Eric O. Callen, considered by most to be the pioneer who began the modern age of human coprolite research (Callen and Cameron, 1955). During the next several decades Callen worked with missionary zeal to convince archaeologists and others about the importance of coprolite research. In retrospect Callen seems like an unlikely person to have become the "father" of coprolite analysis. He spent his professional career as a plant pathologist at McGill University in Canada researching cereal pathogens. His association with coprolites began quite by accident during the 1950s when archaeologist Junius Bird, who had found desiccated human coprolites while excavating the site of Huaca Prieta de Chicama in the coastal region of Peru (Bird, 1948), took them to McGill University in 1951. Bird gave the coprolite samples to T.W.M. Cameron, a parasitologist, and asked if he would be willing to examine them for possible human parasites. Callen, a colleague, teamed up with Cameron in hopes of finding early records of fungal pathogens that might have infected the maize eaten by the early Peruvians. It seems almost humorous in retrospect to realize that the first careful and detailed study of ancient human coprolites was conducted by two nonarchaeologists who were looking for human parasites and traces of maize

Callen's first, and perhaps one of his most significant contributions, was solving the problem of how to reconstitute the desiccated coprolites (Callen and Cameron, 1960). He found that soaking dried coprolites in a weak solution (0.5%) of trisodium phosphate would soften and hydrate samples, allowing for all contents to be separated without harming even the most delicate plant or animal tissues. Applying the trisodium phosphate method to coprolites was important, but equally important was the new set of research standards he set forth for the identification and quantification of coprolite data. The new approach to coprolite research was soon applied to Callen's next projects, including the analysis of coprolites from the Ocampo Caves in Tamaulipas (Callen, 1963, 1965, 1967a) and Tehuacan, Mexico (Callen, 1967b, 1968), from the Neandertal site of Lazaret, France (Callen, 1969), and finally from Pueblo sites in Glen Canyon, Utah (Callen and Martin 1969). It was during the late 1960s that one of us (VMB) corresponded with Callen by mail and spoke with him by telephone on many occasions while conducting a first attempt to process and analyze more than 40 human coprolites recovered from Late Archaic age strata in Conejo Shelter, located near the Rio Grande River in West Texas (Bryant, 1969, 1974a). In April of 1970, VMB first met Callen at the Society for American Archaeology meeting in Mexico City. Two months later, VMB drove to Canada to visit Dr. Callen and to work with him at his lab in MacDonald College of McGill University in Montreal. During the short visit, Callen was flattered that someone interested in coprolites had traveled all the way to Canada to see him, and he was eager to share his techniques and ideas about the potentials of coprolite research. He was also excited about his forthcoming trip that summer to begin work on the coprolites Dr. MacNeish was finding at the Andean site of Pikimachay near Ayacucho, Peru. Both Callen and MacNeish believed that those ancient human coprolites contained clues that would confirm the earliest records of plant cultigen use in South America.

While visiting with Callen in his small lab, which was barely larger than the average-sized bathroom, he expressed a pessimistic view about the acceptance of future coprolite studies and about the apparent minor impact coprolite studies seemed to be having on other fields of science, especially botany and archaeology. He lamented that his colleagues at McGill University considered his work a "waste of time," and that in the decade since he and Cameron had first published their short paper on coprolites (Callen and Cameron, 1960), less than a dozen other researchers worldwide had either collected or tried to examine human coprolites. The reality that even most of these individuals showed little interest in continuing coprolite research also saddened Callen. With Callen's untimely death in Peru during the summer of 1970, while working at the site of Ayacucho (Bryant, 1975), the future of new coprolite studies could

easily have died as well had it not been for a few new advocates who recognized the importance of coprolite data and now took up the challenge that Callen had begun.

From the fifties through the seventies, archaeologists, including J. Richard Ambler, Richard Brooks, Robert Heizer, Cynthia Irwin-Williams, Joel Janetski, Jesse Jennings, Keith Johnson, Richard MacNeish, Don Morris, and Art Rohn realized the importance of saving, curating, and asking specialists to analyze recovered coprolites (Reinhard, 2006). The common theoretical characteristic of these archaeologists was a focus on interdisciplinary scientific analysis and attempts to reconstruct human behavior in an environmental context.

Some of these archaeologists could even be considered coprolite aficionados. Ambler, Heizer, Jennings, Morris, and McNeish focused their excavations around the recovery of coprolites. Jesse Jennings recognized early in his career the importance of science in archaeology. Indeed, his first published article was entitled "The Importance of Scientific Method in Excavation" (Jennings, 1934). Jennings was also one of the first archaeologists to methodically record and collect human coprolites while excavating dry caves in the American West, beginning in 1949. He and his students excavated a number of now famous Utah sites, including Danger Cave, Hogup Cave, Cowboy Cave, and a number of other lesser known shelters and open sites, including some in Glen Canyon, Colorado. His students, Gary F. Fry (Fry, 1969) and Henry J. Hall (Hall, 1969) were the first Utah scientists to analyze human coprolites for dietary information.

J. Richard Ambler excavated Archaic and early agriculture-age coprolites from sites in southern Utah and later found coprolites in rockshelter sites in West Texas. During that time both of us began our careers working with coprolites, Bryant in West Texas and Reinhard in Arizona. Don Morris of the National Park Service noted that "coprolites were the most important data source that he excavated from Antelope House in Canyon de Chelly, Arizona" (D. Morris, pers. commun. to KJR, 1985). Morris excavated Antelope House (in Arizona) during the 1960s and sent a series of coprolites to Texas A & M to be examined (Williams-Dean and Bryant, 1975; Reinhard, 2007).

Robert Heizer excavated sites in the Great Basin of Nevada. He also directed the California Archaeological Survey from 1948 to 1960. Later he was also the Director of the Archaeological Research Facility at The University of California Berkeley from 1960 to 1979. In those capacities, he was able to promote the analysis of coprolites. The initial analysis of coprolites, especially from Lovelock Cave, Nevada, was first published in Science (Heizer and Napton, 1969) and later in other sources (Heizer, 1970). In 1969, three coprolite articles were published in Science. These included Heizer and Napton's article, one by Gary Fry, John Moore and E. Englert (1969), and a third by Fry and Moore (1969). The publication of these three articles in a premier journal during the same year insured that coprolite research would not be forgotten and that that it could offer important biological and cultural windows on the past. A few years later the article by Bryant and Williams-Dean in Scientific American (1975) acquainted not only scientists, but also the general public with the merits and value of examining ancient human fecal remains

Other archaeologists serendipitously became associated with significant coprolite finds at specific sites. Janetski and Johnson discovered coprolites at Antelope Cave in northwestern Arizona (Johnson et al., 2008; Reinhard et al., in press). Richard and Sheilagh Brooks recovered coprolites from La Cueva de los Chiquitos Muertos in Durango, Mexico (Jiménez et al., in press). Irwin-Williams (1980) had the good fortune to uncover thousands of coprolites during her excavations of the Pueblo site of Salmon Ruin, New Mexico (Irwin-Williams and Shelley, 1980; Reed, 2006; Reinhard, 2007; Reinhard and LeRoy-Toren, 2006). Art Rohn directed excavations at Mug House in Mesa Verde Colorado during 1960 and 1961, where he found coprolites. As part of Rohn's effort, he and his group were among the first in the Ancestral Pueblo region to recognize the value of coprolites as keys to the dietary habits of ancient cultures in

that region. Specifically, Rohn and his group noted that prickly pear cactus spines could be seen in human coprolites, and then they went on to experiment by eating prickly pear pads still containing clusters of tiny spines (glochids) to prove that humans could eat those pads with short spines still in place and not suffer any ill-effects (A. Rohn, pers. commun. to KJR, 1986).

In 1968, as part of VMB's dissertation he examined more than 40 human coprolites recovered from Conejo Shelter in Southwest Texas (Bryant, 1969). When Dr. Callen died while working in Peru in 1970, VMB was invited to continue his on-going research. That began VMB's long career over the next 40+ years of continuing the initial work in the field of coprolite analysis and was responsible for shifting the primary focus of coprolite research in North America to his laboratory at Texas A & M University. It has been there that VMB has continued to analyze coprolites and mentor new groups of graduate students wanting to become coprolite experts. These new graduate students have moved coprolite research forward with innovative new ideas while also creating a diversity of new methods for coprolite analyses (Reinhard and Bryant, 2008). A major advantage that helped Texas A & M University become the new center for coprolite analysis was the availability of desiccated fecal remains recovered from a number of rockshelter sites in the arid regions of West Texas and additional coprolites that were being recovered from dry shelters and Pueblo sites throughout the American Southwest. With the site surveys and excavations of West Texas archaeological sites during the WPA (Works Project Association) era of the 1930s, many new sites were first identified and recorded. Tests and preliminary reports of many of those sites would become the focus for later surveys and archaeological excavations more than 30 years later. In addition, the building of Amistad Reservoir, which dammed the Rio Grande River just west of Del Rio, Texas, impounded miles of canyon lands in the Lower Pecos region and flooded over 400 archaeological sites. Before impoundment, however, many of those sites were excavated and their botanical remains, including coprolites, were saved for future analyses. Other sites above the flood pool level of the reservoir were now easily assessable by boat and they also became the focus of an intensive archaeological recovery effort during the 1960s and 1970s. One such site was Hinds Cave, situated in a side canyon leading into the Pecos River and located only a few miles north of the Rio Grande, marking the border between Texas and Mexico

Funded by the National Science Foundation and the National Geographic Society, excavations at Hinds Cave began in 1975 and focused on the recovery of a cultural record for the Lower Pecos region spanning nearly the entire Holocene period. Among the thousands of artifacts and botanical remains were more than 2000 human coprolites recovered from almost every occupational level in the site. Glenna Dean (1978) was the first to examine those and selected 100 of them to analyze for both the macrofossil and microfossil remains. Her initial study inspired others to examine more of the recovered coprolites from the site and to continue building a detailed record of the prehistoric lifestyles and diets of the Lower Pecos region. Janet Stock (1983) completed her graduate work by examining additional coprolites from another time period in the prehistory of Hinds Cave, as did Sherrian Edwards (1990). The preliminary study by Edwards then became the focus of another later study completed by Reinhard and Edwards (Reinhard et al., 2006). Kristin Sobolik (1988) examined coprolites collected in another nearby shelter called Baker Cave and then combined those data with the existing coprolite studies from Hinds Cave and other Lower Pecos sites to chronicle a paleonutrition history of the cultures that once lived in the Lower Pecos Region (Sobolik, 1991, 1994). Danielson (1998, Danielson and Reinhard, 1998) later looked at additional coprolites from Hinds Cave as part of his graduate thesis research. Tim Riley has completed the most recent studies of additional Hinds Cave coprolites (Riley, 2008, 2012). Riley also used the nutritional data from the coprolite studies (Riley, 2012) to predict that the data from these types of coprolite studies will be relevant to our understanding of modern Native American health issues.

In retrospect, after more than 30 years of study of the coprolites from Hinds Cave and other sites in the Lower Pecos region we have obtained a remarkable record of the variety of plant food sources that were exploited and utilized by a series of cultures (Table 1). Using cluster analysis, Riley (2008, 2012) showed that a restricted group of plants formed the Archaic Period's dietary mainstay for peoples in the Lower Pecos region. He found three clusters of plant remains that represent distinctive and important dietary combinations. The first is a combination of agave (Agave lechuguilla), onion bulbs (Allium), and sotol (Dasylirion) with agave dominating. The second cluster is composed of just two plants, prickly pear cactus pads (Opuntia) with secondary consumption of sotol. The third cluster combines prickly pear fruits and some as yet unidentified plant. Thus, of all the plants available throughout a long history of occupation, the dietary mainstay appears to consist primarily of four plants: prickly pear cactus, agave, onion bulbs, and sotol. The reconstructed coprolite dietary data are also complemented by early ethnographic observations, which mention and emphasize the past human dependence upon these key plants (Riley, 2012).

The "Golden Age of Coprolite Analysis" lasted for just over 30 years, beginning in 1960 and ending in the early 1990s (Reinhard, 2006). We believe that the subsequent decline in archaeological coprolite analyses corresponds with the emergence and emphasis on post-processual archaeology in the 1980s and 1990s. Post-processual archaeology, in its many forms, emphasizes subjectivity in archaeological interpretations. Therefore, because coprolite analysis represents the penultimate quantification in terms of archaeological science, it appears to have fallen out of the philosophical importance stressed by the post-processualists. During the two decades of post-processualism, there has been a decline in the number of young archaeologists wanting or willing to train as coprolite analysts. The decline focused around three main reasons. First, there were few places where new graduate students could train to become specialists in coprolite research. Second, and perhaps the major reason, was that not many new archaeological graduate students were willing to study and gain the comprehensive knowledge needed to conduct these types of analyses. Competent research in coprolite research requires studies in a variety of different disciplines including anthropology, archaeology, botany, zoology, parasitology, chemistry, etc. Thus, the prospect of this type of needed training often becomes a daunting obstacle for many. Third, many students find that they will need to spend long and tedious hours looking through a microscope in order to sort and identify the hundreds of tiny plant, animal, and insect remains found in coprolites. For most, that type of future is not nearly as exciting as the potential of working in the field situation surveying or excavating archaeological sites

Despite the tedium of the work and the less than glamorous nature of the discipline, the study of coprolites remains one of the most informative sources available about ancient cultures. When archaeologists excavate site deposits that contain leaves, stems, wood, dried fruits, seeds, fibers, starch grains, phytoliths (plant crystals), or even fossil pollen they often infer that these traces reflect and document the subsistence patterns and food preferences of many past cultures (Sobolik, 2003). However, even though those types of deposits may reflect human cultural lifestyles, those same items might instead have resulted from the activities of carnivores, other animals, wind or water deposits, or a variety of other potential sources, none of which are reflections of human occupation. Thus, the only certainty comes from the careful analysis of human fecal material, which contains the remains of foods that were actually collected and eaten by people.

In coprolite analyses all types of macroscopic and microscopic remains must be counted, tabulated and interpreted in the context of ecological behavior (Reinhard and Bryant, 1992). This requires interdisciplinary training of students skilled in palynology, archaeobotany, zooarchaeology, and parasitology at the very least. In has been our observation (Reinhard and Bryant, 2008) that, although this level of training was willingly pursued by students during the "Golden Age Period,"

TABLE 1. Plant foods eaten in the Lower Pecos region as represented in Hinds Cave and Baker Cave (Reinhard, 1992; Riley, 2012).

Scientific Name	Common Name	Part eaten	Preparation
Agave lechuguilla	agave	caudex (stem base)	Intensive cooking
Allium sp.	wild onion	bulb	various
Amaranthus sp.	pigweed	small fruit	pounding
Brassicaceae	mustard	seed	unknown
Celtis sp.	hackberry	fruit	unknown
Chenopodium sp.	goosefoot	small fruit	pounding
Dasylirion sp.	sotol	caudex (stem base)	Intensive cooking
Diospyros texana	persimmon	fruit	raw
Helianthus sp.	sunflower	achene	cracked, raw
Juglans microcarpa	walnut	nut	cracking
Juniperus sp.	juniper	berries	unknown
Mammillaria	pincushion cactus	fruit	raw
Opuntia sp.	Prickly pear	cladode (pad) and fruit	cooking (pad) raw
			(fruit)
Poaceae	wild grass	caryopsis (seed)	pounding
Polygonum sp.	knotweed	seed	unknown
Prosopis sp.	mesquite	pod	pounding into a
			flour
Vitis sp.	wild grape	fruit	raw
Yucca sp.	Spanish dagger	caudex (stem base)	Intensive cooking

of coprolite studies, since the advent of the Post-processual archaeology period this type of interdisciplinary training is rarely pursued as part of current archaeological training in graduate studies. As evidence, during the past decade only one new graduate student, Riley (2008, 2012), has been willing to commit to this type of work and pursue interdisciplinary coprolite research.

COPROLITE ANALYSIS AND DIABETES RESEARCH

Coprolite analysis should be viewed as a key source of information for researchers interested in the epidemic of Type II diabetes that now affects Native Americans in the southwestern United States (Reinhard et al., in press). However, as of this date, the past analyses of human coprolites from ancient Native American populations has not been assessed for its potential contribution to the debate about the causes of diabetes in these populations, even though there is an extensive literature that has accumulated over the past four decades and which we recently reviewed (Reinhard and Bryant, 2008). We believe that the previous coprolite research and the published results of those analyses can now be presented to test the hypotheses related to the evolution of the diabetes problem among Native Americans living in the American Southwest.

Obesity and Type II diabetes are the most critical health problems now facing Southwestern Native American tribes (Gohdes, 1995; Gohdes and Bennett, 1993; Hill, 1997; CDCP, 2011). These conditions appear to have emerged only during the past 60 years. The "thrifty" gene hypothesis is a prevalent, yet untested, hypothesis that is supposed to explain the emergence of these diseases (Neel, 1962). Researchers believe that the "thrifty" gene promoted good health in ancestral Native American

populations by maintaining an energy balance during "feast or famine" conditions. However, the elimination of "feast or famine" conditions that resulted from the acculturation into modern society combined with declining physical activity patterns, made their "thrifty" gene deleterious. As a consequence, some believe that the thrifty gene is the major cause of up to 50% of the adults in certain Native American tribes being obese and/or diagnosed with Type II diabetics.

Researchers have demonstrated that Native Americans that return to eating a more "traditional" native diet are less likely to develop obesity and/or diabetes (Bellisari, 2008). Defining that original "traditional" diet has been a goal of recent research, both to explain the evolutionary origin of the "thrifty gene" and to develop new methods of dietary intervention for these groups in a clinical setting. However, there is a major gap in the existing knowledge as to exactly what was the traditional diet of many of these cultures. Some researchers suggest turning to the ethnographic record to find clues and then reconstruct those traditional diets (Brand et al., 1990). Perhaps this is a good method for understanding dietary changes during the most recent periods of the past century, but such methods tell us nothing about actual dietary patterns covering the thousands of years the Native Americans have lived and subsisted in the Southwestern regions of America. To try to understand and also reconstruct the dietary habits among early American hunter-gatherers, some researchers have turned to the ethnographic records of other worldwide tribal cultures for examples of what were the types of "traditional" diets (Eaton, 2006). That approach produced a hotly-debated discussion about the accuracy and methods used in the reconstruction of diets (Milton, 2000). Other researchers have tried to integrate a variety of archaeological and bioarchaeological factors into their reconstructions of ancient subsistence patterns (Richards, 2002). Some have even said that changes in cooking patterns and changes in our intake of prebiotic inulintype fructans may be the cause of recent dietary problems (Leach, 2007; Leach and Sobolik, 2010).

Currently, there remains a desperate need for finding a method that can provide empirical details about ancient and traditional diets in the American Southwest, which can be used to test the relevance of the "thrifty" gene and determine if the change from a "feast or famine" condition to current habits is really a primary cause. This gap in knowledge is a problem because it prevents establishing explanations and intervention programs based on an empirical understanding of the evolutionary pressures inherent in the ancestral diets of these Native Americans.

We believe that the gap in direct knowledge of ancient diet patterns can be filled by the past, present, and potential future analysis of ancient human coprolites. Coprolites have been found in archaeological sites in many parts of the American Southwest. Some of these were excavated in the ancestral lands of current tribes. Recently, Reinhard and his colleagues hypothesized that the plant remains from coprolites found at Antelope Cave, Arizona, revealed that ancestral Paiute and Puebloan peoples relied on foods that were very high in fiber, low in fat, and that those foods also had low glycemic indices (Reinhard et al., in press). By looking at various other archaeological sites, which can be linked to modern tribes, the resulting analyses should show which tribes are the most susceptible to a high risk of Type II diabetes and obesity by virtue of their past history of a prolonged reliance on hunter-gatherer types of plant foods. Most importantly, the data will provide basic research for the establishment of dietary intervention recommendations for Native American diabetics.

One recent example comes from the analysis of coprolites recovered from Bighorn Cave where three primary foods were essential elements in the dietary complex of those ancient peoples (Brand et al., 1990; Leach, 2007; Nabhan et al., 1979; Weber et al., 1996, 1998; Williams et al., 2001). One of these three important foods was prickly pear cactus, which we now know decreases blood glucose levels as well the hyperglycemic peak during glucose tolerance testing. In essence, eating prickly pear cactus suppresses sugar metabolism (Becerra-Jiméneza and

Andrade-Cetto, 2011; Gutierrez, 1998). It appears that prickly pear mucilage also slows digestion and thus hinders the sudden rise of blood glucose created by eating foods containing simple sugars. The other two main dietary foods were agave and sotol stem bases, which are full of inulin. Inulin is released slowly into the bloodstream as compared to most typical starches. Thus, like prickly pear cactus, agave can be used effectively in modern treatments for diabetics (Poss et al., 2008). These healthy aspects appear to be related to the low glycemic indices of cactus, agave, and sotol, their high fiber content, and other aspects of their nutritional make up. A mixed diet of these three main plants would inhibit glycemic peaks and energy metabolism. The reliance on these three plants can also be implicated as the foundation for the evolutionary pressure that fixed the "thrifty gene" as being important among prehistoric desert peoples of the American Southwest and their modern descendants today. This could also be applied to the data from Hinds Cave (Table 1).

COPROLITES AND PALEOPATHOLOGY

The most obvious connection of any field with coprolite analysis should have been with paleopathology/bioarchaeology. This is because coprolites reveal the specific disease organisms that caused the nonspecific indicators of disease stress evidenced in bone pathology. This is especially true of vitamin B12 deficiency anemia as represented in the bone pathology of porotic hyperostosis. The long-standing debate regarding the connection of porotic hyperostosis with diets could have been partly resolved by assessing the correlation of parasite prevalence in coprolites with the prevalence of this type of pathology in skeletons. This link was identified and discussed as early as 1992 (Reinhard, 1992, 2006), but relatively few bioarchaeologists recognized the connection between parasites and bone pathology (Walker et al., 2009).

The other reason paleopathologists and bioarchaeologists should promote the study of coprolite research is because parasitology is relatively easy to learn as part of bioarchaeological training. A single course in parasitology would be sufficient to establish a basic expertise in recognizing and diagnosing the presence of a diversity of worm parasites in human coprolites. It is one of the few fields of parasitology in which 90% of regular lab time is spent doing basic microscopic analyses of rehydrated remains. Adding ELISA immunoassay methods would also enable the diagnosis of common protozoan parasites.

Establishing an archaeoparasitology lab is easy and achievable even for anthropology departments with limited budgets. In spite of these apparent benefits, among bioarchaeologists in the United States, only a few, mainly at The University of New Mexico and The University of Indiana, have chosen to do so since 2000.

To demonstrate the relevance of using coprolite data to understand certain ancient diseases, we present an example of how the application of coprolite data can support the most recent hypothesis regarding the causation of porotic hyperostosis (Walker et al., 2009). Walker (2008, p. 119) and his colleagues propose that, "We argue that porotic hyperostosis and many cribra orbitalia lesions are a result of the megaloblastic anemia acquired by nursing infants through the synergistic effects of depleted maternal vitamin B12 reserves and unsanitary living conditions that are conducive to additional nutrient losses from gastrointestinal infections around the time of weaning." They established this conclusion based on an analysis of Puebloan diets and parasitism.

With regard to parasitism, the prehistoric Puebloan parasites have been identified (Table 2). Of the eight parasites known from Puebloan sites, five cause lower vitamin B12 levels. However, of these five parasite types, four are rare and have been found only at Antelope House in Canyon de Chelly, Arizona. These are *Ancylostoma duodenale*, *Strongyloides stercoralis*, *Giardia intestinalis* and *Entamoeba hystolitica*. The most common parasite in Puebloan sites is pinworm, *Enterobius vermicularis*. The prevalence of pinworms in Puebloan sites ranges from 0% to 29% (Reinhard, 1992, 2007, 2008; Reinhard and Araujo, 2008; Reinhard and Bryant, 2008). There is a significant, positive correlation

of pinworm prevalence in coprolites and porotic hyperostosis in skeletons for Puebloan sites (Reinhard, 1992; Reinhard and Bryant, 2008). Normally, pinworms cause very little pathology. However, vitamin B12 malabsorption is one type of pathology that is documented among children infected by pinworms (Cazorla et al., 2006; Sadraei et al., 2007). It is important to note that prehistoric Puebloans had extremely high pinworm infections. As noted by Fugassa et al. (2011) and Jiménez et al. (in press), prehistoric Puebloans had the highest prevalence of pinworm infection of any populations, modern or ancient. In clinical settings, just 5% of fecal samples from pinworm-infected peoples are positive for eggs. Extrapolating from this, a 5% rate of pinworm positive stools indicates that 100% of the people in the source population are probably infected. The prevalence of this type of infection in communities of prehistoric Puebloans ranges to a high of 29%, yet most populations exceeded at least 5%. Therefore, we are justified to predict and infer that at most Puebloan sites pinworms were a chronic and extreme health problem. Therefore, the parasitological data directly support one part of the Walker et al. (2009) model for porotic hyperostosis: that parasite infection contributed to vitamin B12 deficiency. Importantly, the association does show causality between pinworm infection and porotic hyperostosis. This is backed up in a general way by the fact that the more dangerous parasites of the Puebloans also hinder vitamin B12 uptake (Table 2).

Walker et al. (2009) also argue that the diet of Puebloans was deficient in vitamin B12. This assertion is based on isotopic data: "Isotopic studies and other independent lines of evidence indicate that by the time of European contact, 70-90% of the calories in the diets of most Ancestral Puebloans were derived from maize" (Walker et al., 2009, p. 116). This estimate might be closer to the low end since other C4 plants in the diet such as dropseed or amaranth and CAM plants such as prickly pear may have been partly responsible for contributing to the maize signal. The diversity of Puebloan diet has been published in several overview papers (Minnis, 1989, 1991; Reinhard, 1992, 2007) not cited by Walker et al. (2009). However, considering the coprolite data summarized by both Minnis and Reinhard, it is likely that vitamin B12 was also underrepresented in the natural plant diet. Although the plant diet may have been deficient in B12, Puebloans ate significant amounts of meat in the form of small animals, especially rodents and lizards. Summarizing data from Reinhard and colleagues (1992, 2006), the prevalence of small animal residue in Puebloan coprolites includes: 34% among the 96 coprolites examined from Antelope House, AZ, 19% of 16 coprolites examined from Inscription House, AZ, 71% of 35 samples from Chaco Canyon, NM, 23% of 93 coprolites studied from Mesa Verde, CO, and 50% of 30 coprolites examined from Glen Canyon, UT. It is noteworthy that every Puebloan coprolite series exhibits reliance on the protein obtained from hunting small animals. Therefore, even at times when large game was overexploited and unavailable, these ancient groups were able to maintain some meat in the diet through hunting small animals. Therefore, Puebloan diet was usually sufficient in B12, a fact that was missed in Walker's review.

Walker and his colleagues included in their porotic hyperostosis models that focus on the effects of sanitation and starvation. They noted that starvation and failure of sanitation were critical problems affecting Puebloan peoples in prehistory. These effects had previously been demonstrated by coprolite analyses. We had previously described the effects of environmental collapse on diet and disease for Ancestral Puebloans in various case studies, including those of Antelope House and Canyon de Chelly, Arizona (Reinhard and Bryant, 2008). In that work, and based on previous studies (Sutton and Reinhard, 1995; Reinhard and Danielson, 2005), we documented that a long drought forced people to congregate in the areas with surviving water sources found in the region of Canyon de Chelly. These aggregations of people eventually contaminated the water with fecal-borne parasites, including the most debilitating common parasites such as *Ancylostoma duodenale*, *Strongyloides stercoralis*, *Entamoeba histolytica*, and *Giardia intestinalis*. The crowd parasite, pinworm

(Enterobius vermicularis), reached its highest recorded Pueblo prevalence at that site. Diet, as evidenced by phytoliths and cluster analysis of macroscopic remains, focused on the starvation foods documented for Puebloans by Minnis (1991), especially by their reliance on yucca and prickly pear. Not surprisingly, porotic hyperostosis reached its highest levels in Canyon de Chelly.

Walker and his colleagues did an excellent job developing a case for porotic hyperostosis that is related to vitamin B12 deficiency. They summarize that, "We argue that porotic hyperostosis and many cribra orbitalia lesions are a result of the megaloblastic anemia acquired by nursing infants through the synergistic effects of depleted maternal vitamin B12 reserves and unsanitary living conditions that are conducive to additional nutrient losses from gastrointestinal infections around the time of weaning. The Ancient Puebloans also show that such conditions could easily arise among New World agriculturalists lacking foods of animal origin, living in marginal, drought-prone environments in which wild game are easily depleted. Under such conditions, over-exploitation of local animal resources can reach a point where socially disadvantaged people become obligatory vegetarians. Aggregation for defensive purpose would pose additional problems of sanitation, water contamination, and enhanced parasite transmission rates that would further deplete the nutritional stores of affected individuals. Under such conditions, the nutritional deficiencies suffered by mothers would be transmitted to their infants in an exacerbated form" (Walker et al., 2009, p. 119-120).

They based their argument on indirect evidence of diet, isotopic analysis, ethnographic evidence, and clinical data. Although they cited some coprolite references, they managed to miss every relevant coprolite article published between 1992 and 2008, and many dating to the 1980s, which would have been germane to their research. Had they read that body of work, they would have found a basis to support marginal B12 intake from the plant diet, but they would also have learned that the Ancestral Puebloans were never "obligatory vegetarians." Most importantly, they would have learned that the direct drain on vitamin B12 in that population was caused by the parasite population that thrived in the humans that lived in Pueblo communities. They would have discovered a direct and highly significant correlation of parasitism with porotic hyperostosis. Finally, they would have discovered that environmental collapse was linked by coprolite analyses to a decline in diet, a decline in sanitation, and a massive increase in parasitism. All of that information came from the direct evidence of diet and infectious disease as a result of coprolite studies of the remains left behind by the very populations on which their research focused.

This key paper on porotic hyperostosis develops a causal relationship between pathology and a nutrient deficiency. The fact that the authors compiled this information, yet ignored the direct coprolite evidence, is perplexing. It appears that the interdisciplinary environment that fostered the "Golden Age" of coprolite analysis in the latter part of the 20th Century was replaced by barriers between various subfields, especially coprolite studies and paleopathology/bioarchaeology.

THE FUTURE OF COPROLITE ANALYSIS: TURNING THE PAGE

We have made a case that coprolites provide data that fill gaps in our knowledge about prehistoric diet and disease. Once filled, coprolite data can provide a linkage between diet and disease conditions and health resolution. This linkage has been missed by researchers who are exploring both ancient and modern health conditions. Therefore, coprolites are truly the missing links in resolving several current issues in human medicine.

The obscurity of coprolite analysis during the past two decades can be explained by changes in emphases in both biology and archaeology that orphaned the field of coprolite analysis. Postprocessualism in archaeology treated science with cynicism. Coprolite analysis, dependent as it is on multiple lines of investigation, was orphaned as archaeology moved away from scientific methods and theory during the postprocessual period. As a result, few young researchers became interested in or were trained in methods adaptable to the study of coprolites. In the same decades, biology classes on which our training depended fell out of vogue. In the past, palynology, plant anatomy, plant taxonomy, plant ecology, parasitology, and comparative anatomy were core courses for those coprolite specialists who studied archaeopalynology, archaeobotany, archaeoparasitology, and zooarcharology. As molecular biology emerged and then dominated the biological sciences, organismal course offerings such as plant anatomy, plant morphology, phycology, mycology, and others declined at major universities. Therefore, the theoretical basis and the biological methods for coprolite analysis training disappeared from academia in those fields that originally nurtured coprolite analysis during the three decades of the 1960s through the 1990s.

Parasitology has increased its interest in coprolites. Parasitologists in Latin America have long used coprolite data to explore the emergence and control of parasitic disease. In 1995, there were just three parasitologists interested in this area of emphasis. As of this writing, two dozen parasitologists working in Canada, the USA, Peru, Chile, Brazil, and Argentina work in whole or in part with coprolite data. At the University of Nebraska – Lincoln, ten archaeoparasitologists have been trained in the past five years. Thus, the recent focus on coprolites among parasitologists represents an ongoing methodological explosion.

Two controversies have pivoted on evidence obtained from coprolites. First, the Anasazi cannibalism controversy brought coprolites into full view as key evidence for, and against, human cannibalism in the ancient Southwest (Marlar et al., 2000; Lambert et al., 2000; Reinhard, 2006). New biochemical methods were able to show that a prehistoric person ate human muscle tissue (Marlar et al., 2000). However, a review of data from the study of nearly one thousand coprolites in the same Southwestern region showed that this episode of cannibalism was abnormal and could not be explained as being a cultural tradition (Reinhard, 2006). Second, recently coprolites have revealed the presence and timing

TABLE 2. Parasites known from Anasazi sites and the known problems they cause.

Parasite	Symptoms in mother	Symptoms in Fetus	Symptoms in newborn/infant
Acanthocephala	Unknown	Unknown	Unknown
Ancylostoma duodenale	Severe iron deficiency anemia, nutrient malabsorption, lower vitamin B12 levels, alimentary bleeding, hypoproteinemia, fatigue, diarrhea, malnutrition, preeclampsia, death from heart failure in labor	Transplacental infection, abortion, still birth, premature birth	Low birth weight, lower vitamin B12 levels, transmammary infection, severe alimentary bleeding, iron deficiency anemia, nutrient malabsorption, hypoproteinemia, death
Entamoeba histolytica	Iron deficiency anemia, lower vitamin B12 levels, diarrhea, dehydration, shock, death	Reduced intrauterine growth, abortion, still birth, premature birth	Low birth weight, diarrhea, lower vitamin B12 levels,
Enterobius vermicularis	None	None	Lower vitamin B12 levels, minor symptoms
Giardia intestinalis	Diarrhea, malnutrition, lower vitamin B12 levels, anemia, dehydration, nutrient (protein) malabsorption	Reduced intrauterine growth	Low birth weight, lower vitamin B12 levels, malabsorption of iron, diarrhea, malnutrition, nutrient malabsorption
Hymenolepis nana	Minor symptoms	Minor symptoms	Minor symptoms
Strongyloides stercoralis	Hyperinfection, lower vitamin B12 levels, preeclampsia	Unknown	Low birth weight, lower vitamin B12 levels, transmammary infection
Trichuris trichiura	Minor symptoms	Minor symptoms	Rare anemia, rare rectal prolapse

of the first migrants into the New World. The high profile discovery and analysis of coprolites from Paisley Cave, Oregon (Gilbert et al., 2008), representing pre-Clovis humans in the Americas, is again demonstrating the importance of conducting coprolite research.

Forensic science adapts archaeological methods to modern legal situations. Thus, the training of forensic scientists can be based on archaeological remains. This is also giving a boost to coprolites studies, especially coprolites from mummies and burials. As of this writing, 12 students in the University of Nebraska – Lincoln's Forensic Science program have been trained in coprolite analysis of mummies and burials.

The future of coprolite analysis is dependent on capitalizing on the recent high profile discoveries and controversies within archaeology that hopefully will encourage the training of a new generation of coprolite analysts. It is also dependent on allied fields such as parasitology and forensic science. The orphaned status of coprolite analysis in North America will hopefully change soon, as it has in Latin America. In the past decade, coprolite research centers have been established in Peru at the Universidad Peruana Cayetano Heredia; in Brazil at The Brazilian National School of Public Health, at the Oswaldo Cruz Foundation; in Argentina at the National University of Mar del Plata; and in Chile at The Universidad de Tarapacá. These institutions stress and promote the importance of interdisciplinary work on coprolites. We remain optimistic that similar training facilities and programs will be established in archaeology programs in North America, provided science reemerges as the dominant paradigm in archaeology.

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