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Archaeoparasitology of Chaco Canyon

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Undergraduate Thesis Environmental Studies Program University of Nebraska-Lincoln

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ABSTRACT

Ancient cultures of the Colorado Plateau have been a focus of archaeoparasitology since its inception, and a vast parasitological history is recorded in coprolites preserved in this arid region. The inhabitants of Chaco Canyon dominated Ancestral Puebloan culture between 1050 and 1120 AD and were responsible for the construction of great towns, road systems, and early agriculture. Analysis of the parasites preserved in fecal remains contributes to an increased knowledge of ancient Chacoan health and culture. Nineteen coprolites from four sites in Chaco Canyon were rehydrated and analyzed microscopically for parasite remains. Rhabditiform and filariform nematode larvae were found from several sites. Though these larvae closely resemble Ancylostoma duodenale or Strongyloides stercolaris, morphological characters indicate that most likely they were free-living species infesting the feces. Pinworm (*Enterobius vermicularis*) eggs were found at two sites, implying that population aggregation in Chacoan great houses contributed to disease transmission. This study presents the first record of whipworm (*Trichuris* trichiura) among Ancestral Puebloans. Another whipworm from an Archaic site at Chaco Canyon dated to approximately 900 BC represents the first Archaic parasite found on the Colorado Plateau. This species requires warm, moist soil for life cycle completion. Since these conditions are not naturally present on the Colorado Plateau, the presence of T. trichiura can be attributed to an alteration of the environment through irrigation agriculture and the construction of large, communal dwellings. The presence of this species also suggests that the inhabitants of Chaco Canyon had frequent contact with Mesoamerican civilizations.

INTRODUCTION

A thousand years ago, Chaco Canyon was the cultural center of the Southwest. Chaco Canyon is located in the San Juan Basin on the eastern part of the Colorado Plateau in northwest New Mexico. Residents of Chaco Canyon dominated Ancestral Puebloan culture from 1050 to 1120 AD. This society was known for its complex system of multi-storied towns (great houses), roads, and irrigation agriculture (Vivian 1990). Extensive archaeological research of Chaco Canyon has been conducted for the past 150 years, and the area is now preserved as Chaco Culture National Historic Park. This research has created an extensive body of knowledge on Chaco Canyon's early inhabitants.

Archaeoparasitology is the integration of parasitology into archaeological contexts.

Since its emergence in the 1980s, the field has developed in several laboratories around the world, as reviewed by Reinhard (1992). Archaeoparasitology provides a great wealth of information about the antiquity of host-parasite relationships and information on past cultures, including health, diet, trade, migration, and behavior. Its primary sources of data are from coprolites, mummies, and latrine soils. The rehydration and microscopic analysis of any of these types of ancient fecal remains allow well-preserved parasite eggs and larvae to be identified reliably. An understanding of a parasite's life cycle and epidemiology allows inferences to be made concerning its role in a past society.

Ancient cultures of the Colorado Plateau have been a major focus of archaeoparasitology since its inception (Reinhard 1990). The arid Southwest provides ideal conditions for exceptional coprolite preservation, and thus an extensive record of prehistoric New World parasites exists in the region.

The purpose of this study is to determine the parasitological history of the ancient residents of Chaco Canyon, NM. This information will provide new insight into the health and culture of ancient Chacoans and add to the body of knowledge of Archaic and prehistoric New World parasites. The questions addressed include what parasites infected the Archaic and Ancestral Puebloan residents of Chaco Canyon, how infection varied between several sites, and what these infections imply in regard to ancient Chaco culture and health.

The four sites chosen for this study represent differences in time period of occupancy and size of dwelling. Atlatl Cave is a small rockshelter typical of the San Juan Basin and was used by late Archaic hunter-gatherers. Most other Archaic sites from Chaco Canyon are open; here the archaeological record is well preserved by the rock shelter. Carbon dates suggest an occupation of approximately 900 BC (Vivian and Hilpert 2002). Coprolites from an Archaic site in this region have not been examined previously, and they may provide new insight into the nature of Archaic life in Chaco Canyon before the more well-studied Chaco Phenomenon.

Two of the other sites analyzed in this study are Pueblo Alto and Pueblo Bonito, great houses that were in use at the height of Chacoan civilization. Pueblo Bonito began as a small community in 828 AD. By 1050 it had undergone planned expansion into a community of 800 rooms. Construction at the site continued until 1126, leaving it the largest great house in Chaco Canyon (Vivian 1990). Stratigraphic dating for the coprolites analyzed in this study indicates that they were from two periods, 920 to 1020 and 1080 to 1130 (Clary 1984). Pueblo Alto was another large great house where construction began around 1020 and continued through much of the second century. Stratigraphic dating indicates that the Pueblo Alto coprolites used in this study were from 1050-1100. The parasite record from these two locations will be representative of the parasites present at the height of Chacoan culture. Previous analysis of coprolites from

these great houses showed presence of the pinworm, *Enterobius vermicularis*, and nematode larvae that may have been the threadworm, *Strongyloides stercoralis* (Reinhard and Clary 1986).

The final site, Kin Kletso, is a great house built between 1125 and 1130 after the Chacoan culture had largely ended (Vivian 1990). Several coprolites from this site have been analyzed previously, but no parasite remains were observed (Reinhard and Clary 1986). Parasite remains from this site will provide an idea of whether the parasites present in Chacoan villages persisted after the culture's end.

METHODS

Coprolites were obtained from previous archaeological work in Chaco Canyon National Historic Park in Northwestern New Mexico (36°03′ N, 107°57′ W). Nineteen coprolites represent four sites within Chaco Canyon: Atlatl Cave, Pueblo Bonito, Pueblo Alto, and Kin Kletso (Figure 1).

Coprolites were rehyrdated using a method first applied by Callen and Cameron (1960) and reviewed by Reinhard et al. (1986). Depending upon the amount of sample available, 0.1 to 5 g of each coprolite were weighed and rehydrated in 0.5% trisodium phosphate (Na₃PO₄) solution, a mild detergent which effectively dissolves dried fecal material without disfiguring parasite eggs. The coprolites soaked in this solution for 24 to 72 hours. The color of the rehydration fluid was noted at this stage. A dark brown rehydration fluid is indicative of human feces, whereas the feces of other animals produce a lighter-colored fluid (Fry 1985). Ethanol was added to several samples during rehydration to halt bacterial and fungal growth. A portion of each sample was preserved prior to rehydration for future radiocarbon dating to be performed if warranted by the parasites found. Coprolites were examined for evidence of fungal growth and bore holes from arthropods or free-living nematodes prior to rehydration.

Once rehydrated, coprolites were disaggregated with a magnetic stirrer until all fragments were broken. At this time a *Lycopodium* tablet dissolved in aqueous hydrochloric acid (HCl) was added to each sample. Each tablet contains a known number of club moss spores that will appear in the same fraction of the rehydrated coprolite as parasite remains. This allows a quantitative comparison of parasites from different samples. The resulting solution was poured through a geological screen, and the solution that passed through the screen was centrifuged to concentrate microscopic remains, including parasites.

One sample was processed with zinc bromide $(ZnBr_2)$ to separate microscopic remains from sand. The sample was centrifuged with $ZnBr_2$ until it separated into light and heavy fractions. Both were centrifuged until the supernatant ran clear, and the light fraction was used in subsequent microscopic analysis.

Processed material was preserved in glass vials with 70% ethanol. Solution was transferred to 25x75 mm glass slides with a glass pipette and mounted in glycerine. This end result is essentially a fecal smear; parasite eggs are identifiable by scanning with light microscopy.

Once potential parasitological remains were found, preliminary identifications were made by morphological characters and micrometer measurement. Independent confirmation was sought from parasitologists in several labs including the HW Manter Lab at the University of Nebraska-Lincoln, the Paleoparasitology Lab at the Brazilian School of Public Health (FIOCRUZ), and the Paleoparasitology Lab at Universidad Nacional de Mar del Plata, Argentina. All parasite remains were photographed.

RESULTS

Coprolites did not show evidence of infestation by arthropods or free-living nematodes.

One coprolite from Atlatl Cave was suspected to be geological during rehydration; this was confirmed by microscopy. Rehydration fluid color and microscopic analysis indicated a human origin for all other samples. No parasite remains were observed in the coprolites from Pueblo Alto.

Two coprolites were positive for *Trichuris trichiura* eggs (Figure 2), one from Atlatl Cave (24 eggs/gram of coprolite) and one from Pueblo Bonito (29 eggs/gram). This represents the first record of *T. trichiura* among Ancestral Puebloans and the first parasite record for an Archaic site on the Colorado Plateau.

Two samples were positive for *Enterobius vermicularis* (Figure 3), one from Pueblo Bonito (16 eggs/gram; 10% prevalence, n=10) and one from Kin Kletso (106 eggs/gram; 25% prevalence, n=4). This is the first parasite record for Kin Kletso.

A first stage rhabditiform larva was also found in the Atlatl Cave sample positive for *T. trichiura* (Figure 4). This larva was preserved poorly, so it cannot be determined with certainty whether it was of a parasitic or free-living species.

Two samples from Pueblo Bonito contained rhabditiform larvae (Figures 5 and 6), and one also contained a filariform larva (Figure 7). These had parasite counts of 467 and 2,008 larvae per gram of coprolite. Morphological characteristics suggest that these larvae were of a free-living nematode species that infested the feces.

DISCUSSION

The presence of *T. trichiura* at Atlatl Cave and Pueblo Bonito represents both the first Archaic parasite found on the Colorado Plateau and the first record of the whipworm among Ancestral Puebloans. Eggs of *T. trichiura* have been found previously at Elden Pueblo, AZ (Reinhard et al. 1987), but this species is otherwise absent from the parasite record of ancient Southwestern cultures. Trade and seasonal migration to the Verde River Valley of the Sonoran Desert were suggested to be the source of whipworm infection for Elden Pueblo.

Archaic and prehistoric residents of Chaco Canyon incorporated many small mammals into their diets, including the prehistoric woodrat (Clary 1984). Woodrats are host to *Trichuris muris*, whose eggs are similar in appearance to *T. trichiura* but slightly larger. Confalonieri et al. (1985) demonstrated that ancient *Trichuris* eggs do not significantly change in shape or size when dessicated. Thus the measurements of several *Trichuris* species made by Thienpont et al. (1979) can be used to identify the *Trichuris* eggs found in these samples as those of *T. trichiura*.

T. trichiura requires warm, moist, shaded soil for egg embryonation (Roberts and Janovy 2009). These conditions are not naturally present on the arid Colorado Plateau, so the presence of this species in ancient times indicates that the inhabitants of these sites altered the environment in such a way that allowed transmission to occur. Recent analysis of maize pollen from Chaco Canyon indicates that the Archaic occupants of Atlatl Cave around 900 BC were early maize agriculturalists (Hall 2010). Thus the presence of whipworm in an Archaic site can be at least partially explained by the shift toward agriculture from a solely hunter-gatherer lifestyle. As Puebloan culture developed, irrigation systems and human population density grew; Chaco Canyon shows the highest complexity of irrigation known for Ancestral Puebloans and near-urban conditions in great houses (Vivian 1990). Thus the persistence of T. trichiura into

Puebloan culture and its occurrence in the canyon's largest great house correspond to these changes.

These findings are consistent with previous hypotheses about the impacts of a sedentary, agricultural lifestyle on patterns of infection. Cockburn (1971) proposed that the evolution of parasitism followed the evolution of hosts and their culture. This is evidenced by previous parasitological data from the ancient Colorado Plateau showing that occasional infections present in hunter-gatherers became much more severe among agricultural peoples (Reinhard 1988).

The presence of *T. trichiura* at Chaco Canyon is also significant to the question of its arrival to the region. There has been a long-held consensus among many archaeologists that prehistoric cultures of the Southwest, including the Ancestral Pueblo culture of Chaco Canyon, developed independently of Mesoamerican influence. However, material items recovered from Chacoan sites, such as copper bells, macaws, and certain styles of pottery suggest direct involvement with Mesoamerican cultures (Vivian 1990). Washburn et al. (2011) recently reported the presence of theobromine, a biomarker for cacao, in Chacoan drinking vessels. Cacao is native to Mesoamerica, so this finding also suggests frequent trade between Ancestral Puebloans of the Southwest and Mesoamerican states. The presence of *T. trichiura* at Chaco Canyon corroborates hypotheses for contact with Mesoamerica where warm, moist soil allows transmission.

The presence of this species at Chaco Canyon is interesting in another regard. *T. trichiura* is an heirloom parasite, meaning that it evolved along with its human hosts. The arrival of this species to the Americas has been the subject of a long debate, as reviewed by Araújo et al. (2008). It was long believed that the species arrived to the New World with European colonization, but there are now numerous records of pre-Columbian infection in the Americas.

Because the life cycle of this species dictates that it cannot survive in especially cold environments, it is unlikely that it arrived to the New World with hosts traveling across Beringia. Thus, this record of *T. trichiura* in Archaic and prehistoric America supports hypotheses for alternate routes of early human contact with the continent.

The human pinworm, *Enterobius vermicularis*, is an extremely common parasite in ancient and modern populations alike, and it appears at a very high prevalence in several previous parasitological records of Ancestral Puebloans. Previous analysis of coprolites from Chaco Canyon demonstrated a 20% prevalence of *E. vermicularis* at Pueblo Bonito (Reinhard and Clary 1986). Other Puebloan sites have shown even higher rates of infection, up to 29% at Antelope House. In a clinical setting, only 5% of the feces from pinworm-infected people test positive for pinworm eggs, so these records are indicative of a very high prevalence of pinworm infection (Reinhard 2008).

In the present study, Pueblo Bonito and Kin Kletso had pinworm prevalences of 10% and 25%, respectively. These prevalence values are indicative of a fairly high level of infection, though the low egg counts within infected coprolites is puzzling. Previous rehydration of coprolites for parasite analysis may have been performed with hydrofluoric acid (HF) to separate parasite remains from sediments. It is possible that more pinworm eggs are present in these coprolites and could be isolated with HF or ZnBr₂ (Reinhard et al. 1986).

Pinworm is generally not pathogenic, so its presence alone is not especially significant in determining the health of Ancestral Puebloans. However, pinworm is transferred by person-to-person and environmental routes, with increased transmission in conditions of cramped living or poor hygiene (Roberts and Janovy 2009). The presence of pinworm is a good indicator for the conditions of ancient hygiene and the potential presence of other diseases with similar

transmission patterns, such as tuberculosis (Reinhard 2008). Pinworm is a crowd disease, and its prevalence reflects the extent to which prehistoric people were aggregated. By the 1100s, Chacoan great houses stood up to 4 stories tall, with up to 500 rooms and 30 ceremonial kivas (Vivian 1990). Thus Chacoan society was based on near-urban conditions of population aggregation that were highly suitable for pinworm transmission.

Possibly the greatest source of confusion in parasitological analysis of coprolites is distinguishing parasitic helminths from free-living species (Reinhard et al. 1986). It is important to consider the possibility that any larval nematode found in fecal material could have been free-living, as numerous free-living species commonly infest dung. Some larvae of these free-living species closely resemble common human parasites, such as the hookworm, *Ancylostoma duodenale*, and threadworm, *Strongyloides stercoralis*. This similarity is especially problematic when larvae are poorly preserved or desiccated, such as some specimens found in this study.

Some morphological characters suggest that the larvae recovered from Atlatl Cave and Pueblo Bonito were of *A. duodenale* or *S. stercoralis;* the presence of either of these species at Chaco Canyon would fit with the presence of *T. trichiura*. Like the whipworm, these species require warm, moist soil for life-cycle completion and would suggest Mesoamerican contact and environmental alteration (Roberts and Janovy 2009).

Several diagnostic characters are useful in determining the nature of the helminths present in coprolites. Normally, a wide range of life-history stages are present in feces if worms were free-living. The presence of only first-stage larvae seen here is more typical of parasitic life-cycles, especially *S. stercoralis* (Reinhard et al. 1986). Additionally, dry feces do not provide a hospitable environment for free-living nematodes. The lack of evidence of arthropod

infestation or fungal growth seen in these coprolites suggests that they desiccated quickly without adequate time for infestation by free-living nematodes.

Previous examination of coprolites from Pueblo Bonito produced both free-living larvae and some suggested to be *S. stercoralis*. The morphology of these larvae resembled the first or second stages of juvenile *S. stercoralis* or *Rhabditis*, a free-living nematode (Reinhard and Clary 1986). The larvae recovered from Pueblo Bonito in the present study were very similar in size and morphology.

Despite the considerations above and the resemblance of these larvae to certain stages of both *A. duodenale* and *S. stercoralis*, it is likely that the larvae from both the previous and present analyses were from a free-living species. Though poor preservation prevented all larval characters from being viewed, overall morphology suggests that they were free-living.

The esophagus-intestine juncture is located at a third of the total body length, a characteristic of *S. stercoralis* rhabditiform larvae. However, the curved intestine and presence of a cuticle do not suggest *S. stercoralis*. The larvae are too short to be *A. duodenale*, and tail morphology is not consistent with this species. Though Reinhard and Clary (1986) suggested that the similar larvae observed in their study might be of *S. stercoralis*, they were probably of the same free-living nematode seen here. The rhabditiform larva from Atlatl Cave does not demonstrate enough morphological characters to confirm a parasitic origin. Therefore, for the Colorado Plateau, confirmed diagnoses of *A. duodenale* and *S. stercoralis* exist only for the Antelope House Pueblo in Canyon de Chelly, AZ (Reinhard and Bryant 2008).

The difficulties encountered in identifying these larvae reiterate the challenges of larval helminth coprology and the need to seek verification for any potential parasite remains found in ancient samples.

Chaco Canyon was significant to the prehistoric Southwest as the center of Ancestral Puebloan culture, and its ancient residents have received great attention by archaeologists.

Analysis of coprolites for ancient parasite remains brings unique insight into many aspects of ancient Chacoan health and culture, including trade, migration, sedentism, agriculture, and population aggregation. High pinworm prevalences imply that populations in Chacoan great houses were highly aggregated and lived in conditions of poor hygiene. This study presents the first parasite record for an Archaic site on the Colorado Plateau and the first whipworm record for Ancestral Puebloans. These finds correspond to the arrival of maize agriculture to the region, imply regular contact with Mesoamerican cultures, and suggest an alteration of the environment through irrigation and the construction of large, communal buildings.

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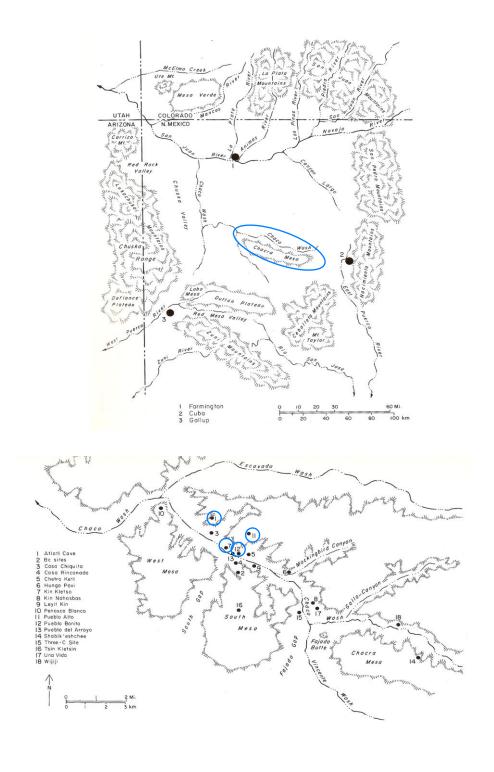
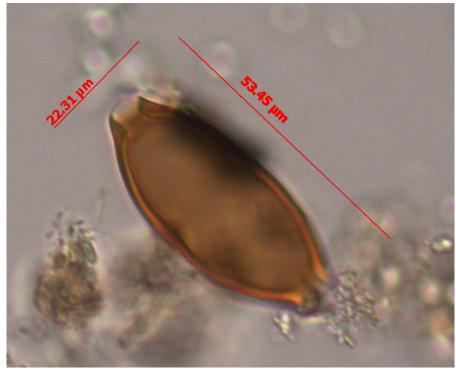


Figure 1. Chaco Canyon is located in northwestern New Mexico (top). Coprolites were analyzed from four sites in the canyon (bottom): Atlatl Cave, Pueblo Alto, Pueblo Bonito, and Kin Kletso (After Vivian 1990).



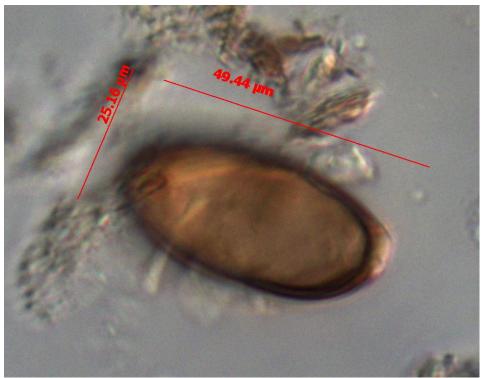


Figure 2. *Trichuris trichiura* eggs from Atlatl Cave (top) and Pueblo Bonito (bottom). These eggs fell within the expected size range for *T. trichiura* and well below that of *T. muris*.



Figure 3. Poorly preserved *Enterobius vermicularis* egg with visible larva from Pueblo Bonito.

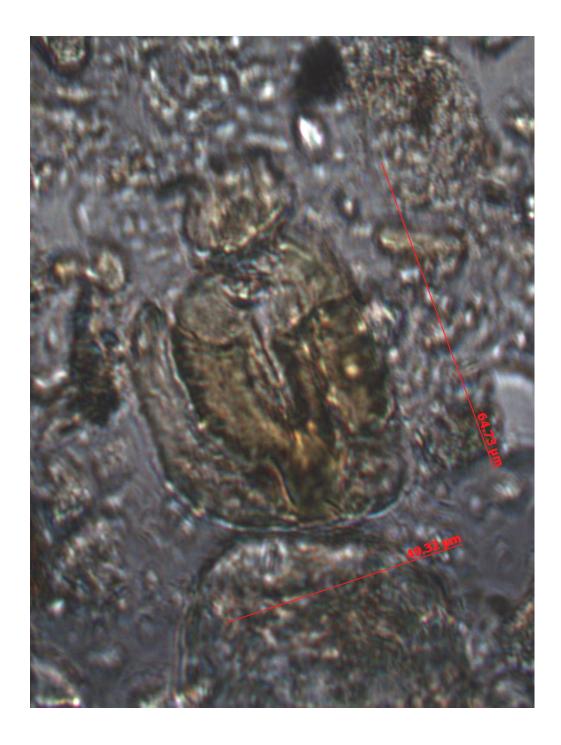
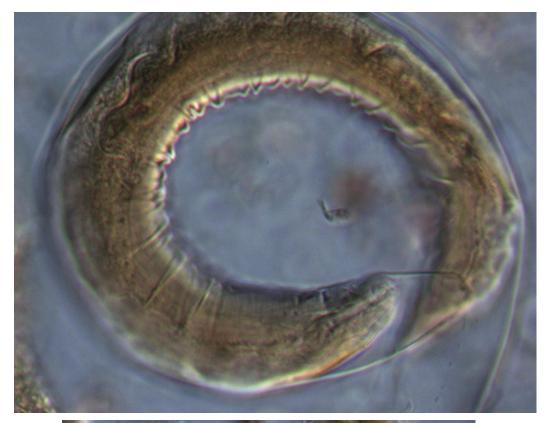


Figure 4. Poorly preserved larval nematode from Atlatl Cave. Morphologically, this larva resembles a juvenile (L1) stage of *A. duodenale*, but the overall dimensions are too small to be of this species.





Figure 5. Rhabditiform larvae from Pueblo Bonito. These larvae shrank within their cuticles during desiccation and resemble those observed by Reinhard and Clary (1986). They are believed to be of a free-living species. The esophagus-intestine juncture (marked by arrow) is located at a third of the total body length, a characteristic of *S. stercoralis* rhabditiform larvae. However, the curved intestine and presence of a cuticle do not suggest *S. stercoralis*. The larvae are too short to be *A. duodenale*, and tail morphology is not consistent with this species. Note that the structure in the top worm resembling an oxyurid vulva is merely a cuticular fold.



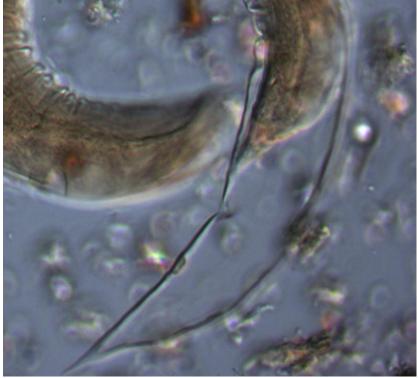


Figure 6. The mouth and curved intestinal tract (top) and pointed tail (bottom) are clearly visible in this rhabditiform larva from Pueblo Bonito. The bottom image illustrates the degree to which the desiccated larvae shrank within their cuticles.



Figure 7. Filariform larva from Pueblo Bonito. This larva resembles the infective stage (L3) of *A. duodenale*. However, the curved intestine and body length are probably indicative of a free-living species.