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PARASITISM OF PREHISTORIC HUMANS AND COMPANION ANIMALS FROM ANTELOPE CAVE, MOJAVE COUNTY, NORTHWEST ARIZONA

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1 ABSTRACT: Previously, we reported a tick recovered from Antelope Cave in extreme northwest Arizona. Further analyses of coprolites from Antelope Cave revealed additional parasitological data from coprolites of both human and canid origin. A second tick was found. This site is the only archaeological locality where ticks have been recovered. We also discovered an acanthocephalan in association with *Enterobius vermicularis* eggs in the same coprolite. This association shows that the coprolite was deposited by a human. This discovery expands our knowledge of the range of prehistoric acanthocephalan infection. In addition, findings from canid coprolites of *Trichuris vulpis* are reported. This is the first published discovery of *T. vulpis* from a North American archaeological context. The close association of dogs with humans at Ancestral Puebloan (Anasazi) sites raises the potential that zoonotic parasites were transferred to the human population. The archaeological occupation is associated with the Ancestral Pueblo culture 1,100 yr ago.

Antelope Cave is the most important site related to understanding the range of Ancestral Pueblo (Anasazi) behaviors that defined ancient parasitism in the southwestern Unites States. Previous studies have focused on Pueblo sites in the Colorado Plateau core area (Reinhard et al., 1987; Reinhard, 1990). Relative to previously studied sites, Antelope Cave is located in the northwest corner of Arizona, within the Great Basin cultural area. It represents Ancestral Pueblo adaptation in its extreme cultural and environmental range.

Ancestral Pueblo people and their domesticated dogs were adapted to a diverse range of environments in Arizona, Colorado, Utah, and New Mexico. Dogs were present in the Southwest throughout prehistory (Colton, 1970). At least 1 zoonotic dog parasite has been identified. *Strongyloides stercoralis* was found in dog coprolites at the northeastern Arizona site Antelope House in Canyon de Chelly, Apache County (Reinhard, 1985, 1990; Reinhard et al., 1987). We investigated human and dog parasites at Antelope Cave in Arizona's northwestern corner. Excavations at this site recovered coprolites of both humans and dogs.

A more important aspect of Antelope Cave is the fact that the people in this area subscribed to a pattern of subsistence that resulted in exposure to infections with thorny-headed worms (Phylum: Acanthocephala). On the margins of the Great Basin, Ancestral Puebloans adapted to relatively scarce resources by collecting and eating a wider diversity of wild foods. Several authors, e.g., Fry (1977), Madsen and Kirkman (1988), and Madsen (1989), have shown that insects were an important food source in the area within the Great Basin cultural area. Sutton (1988) summarized the ethnographic record of insect consumption for Great Basin Tribes. He calculated the nutritional value of insects referred to by ethnographic records and concluded that insects were a critical dietary resource rather than an occasional food item. The insects eaten in the region in historic times included grasshoppers, crickets, cicadas, shore flies, sphinx moths, pandora moths, bee larvae, and June beetles. This well-documented dietary practice overlaps the region where prehistoric acanthocephalan infections were reported (Fry, 1977). This overlap presents a convincing paleoepidemiological picture of the relation of subsistence to thorny-headed worm infection.

One of the truly fascinating aspects of archaeoparasitology is the evidence of zoonoses that can be revealed by studies of ancient peoples (Reinhard and Bryant, 2008; Sianto et al., 2009). The most long-standing zoonosis topic is the acanthocephalan parasitism of Archaic hunter-gatherer peoples in the Great Basin and its margins (Moore et al., 1969; Hall, 1972, 1977; Fry, 1977; Gonçalves et al., 2003; Reinhard and Bryant, 2008). Coprolite analysts have identified acanthocephalan eggs from sites through 10,000 yr of time (Table I).

Acanthocephalan infection was a focus of research early in the history of archaeoparasitology (Moore et al., 1969), and these first findings made a sensation among acanthocephalan experts (e.g., Schmidt, 1971). As coprolite analysis expanded from the Great Basin to other regions of the Southwest, it became obvious that prehistoric acanthocephalan infection was largely limited to the Great Basin cultural region, both in the geographical Great Basin per se, and on its eastern margin (Reinhard, 1990). The margins include Great Basin borders with the Colorado Plateau and the Uinkaret Plateau. The extensive analyses of coprolites from the Ancestral Pueblo core region revealed no evidence of infection, with the exception of a single find mentioned by Gummerman et al. (1972) from Black Mesa, Arizona. However, this discovery was not fully documented and remains anecdotal to this day.

Some finds of acanthocephalans from archaeological sites were identified as *Moniliformis clarki* (Moore et al., 1969; Fry, 1977). There are clinical reports of human infection with a related species in Nigeria and Iran. Ikeh et al. (1992) found *M. moniliformis* eggs in the feces of a man with intestinal symptoms. Also, *M. moniliformis* was found in feces of a child (Sahar et al., 2006), while Berenji et al. (2007) reported *moniliformis* in Iran from a 2-yr-old girl who ate cockroaches. Her symptoms were abdominal pain, diarrhea, vomiting, and facial edema. Salehabadi et al. (2008) also reported *M. moniliformis*. Thus, the archaeology reports, combined with current clinical reports, show that both species of *Moniliformis* can infect humans. Experimental infection with Archiacanthocephala in humans causes severe abdominal pain,

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TABLE I. Previous acanthocephalan finds from the Great Basin and Colorado Plateau.

Site	No. coprolites analyzed	No. positive	Date range (yr ago)
Great Basin			
Danger Cave, Utah	46	6	11,500–1,800
Dirty Shame, Oregon	13	1	9,500–5,900
Hogup Cave, Utah	50	2	10,000–4,000
Colorado Plateau			
Clyde's Cavern, Utah	25	2	1,560–500
Glenn Canyon, Utah	30	2	900–700

diarrhea, sleepiness, and tinnitus in the ears (Moore et al., 1969). Natural infections in humans cause ulceration in the abdominal wall and intensive pain (Leng et al., 1983).

The picture of acanthocephalan parasitism for the Great Basin became more complicated when Reinhard (1990) determined that at least 2 additional morphological types of eggs were present, beyond those reported from Danger Cave and Hogup Cave, Utah. Those from Danger Cave and Hogup Cave were consistent with *Moniliformis clarki*, while the eggs from Clyde's Cavern, Utah, and Dirty Shame Rockshelter, Oregon, represented 2 distinct morphologies. The authors of these studies did not, however, provide sufficient details of the finds to allow for diagnosis (Reinhard, 1990). Therefore, it appears that as many as 3 species of acanthocephalans infected prehistoric people in the Great Basin.

Previously, we reported the discovery of a tick from Antelope Cave in Mojave County, Arizona, on the Uinkaret Plateau (Johnson et al., 2008). With regard to physical geography, Antelope Cave is outside of the Great Basin. However, with regard to cultural geography, the Kaibab and Piaute Tribes are considered part of the Great Basin cultural geography. The cultural affinity of the Uinkaret region to the Great Basin suggested to us that prehistoric people there may have been infected with acanthocephalans. Therefore, we undertook the analysis of coprolites to test for infection.

MATERIALS AND METHODS

Antelope Cave is located on the Arizona strip in the northwest corner of Arizona, 40 km southeast of the nearest city, St. George, Utah. It is a large, subterranean limestone cavern sunk into the semiarid, gently rolling plains of the Uinkaret Plateau, about 1,420 m above sea level. The cave interior measures 107 m north-south and 46 m east-west. Prehistoric Native Americans first occupied this underground site 4,000 yr ago and made use of it at least until A.D. 1150. The most abundant cultural materials at the cave are attributed to the Ancestral Puebloan (Virgin Anasazi) peoples. Lyneis (1995) characterized the Virgin Anasazi as typically organized into small communities that practiced agriculture (corn, beans, and squash) along with hunting game and gathering wild plant foods. Some culture contact with adjacent societies is evident from the occurrence of Fremont (Utah) pottery in Virgin Anasazi sites, but more intense relations appear to have been with the Kayenta Anasazi in northeastern Arizona.

Scientific investigations at Antelope Cave began in 1954 under the direction of Robert Euler of the Museum of Northern Arizona. Subsequent excavations were carried out by archaeologists from the University of California–Los Angeles (UCLA) (Johnson and Pendergast, 1960) and Brigham Young University (Janetski and Hall, 1983; Janetski and Wilde, 1989). The present report focuses on the analysis of human and animal coprolites from the UCLA excavations.

In the summer of 1959, UCLA archaeologists excavated five 2 × 2-m
2 pits into the dry, prehistoric midden deposit of Antelope Cave. These units were designated AC59-1 through 5. The midden deposit was greatest in adjoining units AC59-2 and 5, where the depth of the cultural debris measured 183 cm from the surface of the ground to the bottom of the pits. Several ^{14}C dates indicate that the midden was deposited from A.D. 680 to A.D. 960. Thus, the Virgin Anasazi people successfully occupied Antelope Cave, perhaps intermittently, for more than 250 yr.

The 5 excavation units yielded a rich assortment of cultural materials. Artifacts include fiber sandals, cordage, basketry, rabbit nets, feather ornaments, wooden arrows, pottery sherds, lithic seed grinding tools, and more.

Factors of special interest are the animal bones recovered during the excavation. Jacob Fisher at the University of Washington has analyzed faunal remains that number 23,400 specimens. Of these, all but 300 represent jackrabbits or cottontail rabbits. The following is a list of the identified animals in the faunal collection: mountain sheep (Artiodactyla); bats (Chiroptera); wood rats and pocket gophers (Rodentia); jackrabbits and cottontail rabbits (Lagomorpha); dogs (or foxes) and bobcats (Carnivora); owls, ravens, flickers, and ducks or geese (Aves); and turtles (Testudines).

Additional Antelope Cave fauna identified in the Brigham Young University collections include mule deer, antelope, wild turkey, mouse, Canada goose, pinyon jay, and thresher (Janetski and Hall, 1983; Janetski and Wilde, 1989). From a parasitological perspective, it is important to emphasize that peccary, pig, and raccoon were not present in the cave. If present, these animals could have hosted acanthocephalans.

There were 190 coprolites, both human and animal, recovered by UCLA in 1959. These were not concentrated in latrine areas, but they were found scattered throughout all 5 pits. The highest concentration of prehistoric feces, 13, was encountered in the 76–91 cm level of AC59-4.

Here, our main concern is with coprolite AC643. It came from the top 0–15 cm level in excavation unit AC59-2 in Antelope Cave. A yucca quid from this level yielded a ^{14}C date of 1,230 ± 40 yr B.P., cal A.D. 680–890 (Beta 257786).

Since the publication of Johnson et al. (2008), we have analyzed an additional 22 human and 4 canid coprolites for parasite evidence. The coprolites were analyzed in 3 different laboratories. Samples of 21 coprolites were separated and sent to laboratories in Brazil and Argentina. All samples in Brazil and Argentina were examined using the same rehydration technique (Callen and Cameron, 1960), and then they were preserved in a 5% formalin/acetic solution. Slides were prepared after spontaneous sedimentation (Lutz, 1919) in conical glass jars and passed through a 297-μm-diameter screen. Sediment obtained was used to prepare 10 slides for each coprolite, which were observed using a microscope at ×100, and pictures were taken at ×400. The analysis in Nebraska followed the procedures preciously published by Johnson et al. (2008), and the samples are stored in vials in glycerine at the Harold Manter Laboratory, University of Nebraska State Museum, Lincoln, Nebraska.

RESULTS

The results of the parasitological analysis are presented in Table II. One of the samples examined (AC643) was positive for

TABLE II. Parasitological findings from Antelope Cave.

Number analyzed	Number positive	Parasites observed
Human coprolites		
22	2	*Dermacentor andersoni* only C1
22	1	*Macracanthorhynchus ingens* cf., and *Enterobius vermicularis*
22	4	*Enterobius vermicularis* only
Animal coprolites		
4	1	*Trichuris vulpis* only
4	1	*Trichuris vulpis* and unidentified egg

FIGURE 1. Thorny-headed worm egg (Acanthocephala). The egg is consistent with species of *Macrocanthorhynchus*. Bar = 20 μm.

Enterobius vermicularis. In this same sample, thick-shelled eggs were also found. The general morphological characters and the presence of characteristic hooks in 1 extremity of the cystacanth led to the diagnosis of Acanthocephala (Fig. 1). Ten acanthocephalan eggs without external shells were measured. The external shells did not preserve well enough for measurement. The dimensions of the eggs (without the outer shell) were 30.0–37.5 (35.62 ± 2.92; n = 6) μm × 50.0–57.5 (55.62 ± 2.82; n = 6) μm. 3 4

The poor preservation of the outer shells complicated identification. Two species, *M. ingens* and *M. hirudinaceus*, match the morphology of the recovered eggs. The dimensions of the eggs are 5 closest to the lower limit of *M. hirudinaceus* eggs (Table III). The size of *M. dubius* eggs, based on measurement of the inner shell, is greater than that of the Antelope Cave specimen, also based on inner shell measurement. Therefore, the archaeological eggs are more consistent with *M. hirudinaceus*. Other characters support the *Macracanthorhynchus* diagnosis. *Macracanthorhynchus* spp. eggs have a dark color, while *Moniliformis* spp. eggs are translucent. *Macracanthorhynchus* spp. eggs possess a raphe on the outer shell. *Moniliformis* species have 2 embryonaric membranes, while the *M. hirudinaceus* acanthor is covered by 4 membranes (Soulsby, 1987), and the second membrane is dark brown (Soulsby, 1987). These characters of *Macracanthorhynchus* spp. are consistent with the archaeological eggs (Fig. 1). Moreover, the acanthor is generally more elliptical in species of *Moniliformis* than in *Macracanthorhynchus* spp.

According to Taylor et al. (2001), the archeacanthocephalan species, i.e., *Acanthocephalus rauschi*, *M. hirudinaceus, M. ingens*, and *Moniliformis moniliformis*, are all zoonotic. The *M. hirudinaceus* life cycle includes a vertebrate host, mainly wild and domestic pigs, which are infected by eating beetles (Miyazaki, 1991). There are also records of human infections by paleoacanthocephalans, i.e., *Bolbosoma* sp. (Tada et al., 1983), *Corynosoma* sp., and *Acanthocephalus* sp. (Schmidt, 1971; Cabrera et al., 1999).

Lambl first recorded human infection by *M. hirudinaceus* in 1959, in Prague, Czech Republic (Schmidt, 1971). There have been other findings of adults of this parasite in humans, and eggs have also been found in fecal material (Gonzaga, 1921). However, adults and eggs have never been found together in the same human individual. Therefore, true infection and complete adaptation of the parasite to humans has never been proven.

TABLE III. Summary of the egg measurements of each of the different modern acanthocephalan species. Measurements are in μm. Archaeological specimens are susceptible to shrinkage, especially with regard to the outer shell. This makes detailed comparison to modern eggs impossible. The inner shells of the archaeological specimens are well preserved, but few earlier authors measured the inner shells. 6

Species	Inner shell width (μm)	External shell width (μm)	Inner shell length (μm)	External shell length (μm)
Antelope Cave	33–39	30.0–37.5	53–58	50.0–57.5
M. hirudinaceus Miyazaki (1991)		40–50		80–100
M. hirudinaceus Acha and Szyfres (1989)				70–100
M. hirudinaceus Olsen (1977)		46–65		70–100
M. hirudinaceus Travassos (1917)		51–56		92–100
M. hirudinaceus Soulsby (1980)		40–65		67–110
M. moniliformis Sahar et al. (2006)		65		100
M. moniliformis Al-Rawas et al. (1977)		41–70		80–102
M. moniliformis Neafie and Marty (1993)		54		103
M. dubius Miyazaki (1991)	37–43	57–67	88–97	109–123
M. dubius Olsen (1977)		54–64		112–120
M. dubius Sahba et al. (1970)		41–54		70–83
M. dubius Yamaguti (1963) in Sahba et al. (1970)		40–63		85–163
M. dubius Travassos (1917)		71–74		124–127
O. martini Schmidt (1977)		40–44		60–64
O. schacheri Schmidt (1972)		40–44		70–78
O. oncicola Travassos (1917)		71–75		99
O. canis Soulsby (1980)		40–50		59–71

C2 C3

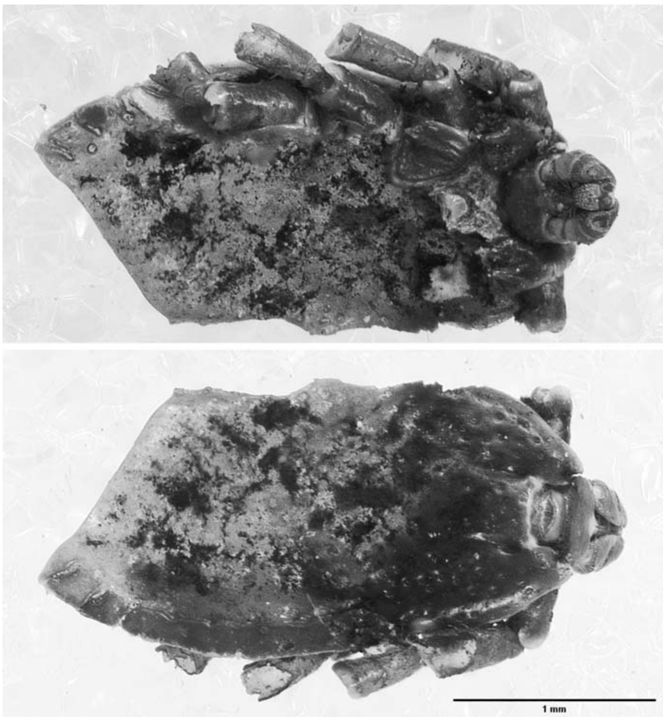

FIGURE 2. The tick recovered in this analysis is very similar to the specimen of *Dermacentor andersoni* recovered from Antelope Cave and presented by Johnson et al. (2008). The upper image is the ventral view, and the lower image is the dorsal view. The capitulum is visible from both sides. The coxal spurs are obvious, and palpi are short, about as long as the basis capituli. The basis capituli is essentially rectangular. Unlike the previously published specimen, the hypostome is well preserved in this specimen.

Although species of *Macracanthorhynchus* are globally distributed, modern human cases have higher prevalence in China due to consumption of raw arthropods as food or medicine (Miyazaki, 1991).

Macracanthorhynchus ingens is endemic to Arizona, but there is only 1 clinical record of a *M. ingens* human infection. This was in a 10-mo-old child in Texas who became infected by eating crickets (Dingley and Beaver, 1985). Millipedes and insects, such as roaches and crickets, are the intermediate hosts of *M. ingens*.

DISCUSSION

The Antelope Cave find adds even more complexity to our knowledge of zoonotic parasitism by acanthocephalans. First, this is the first find of acanthocephalan infection in Arizona and within the Ancestral Pueblo cultural range. Second, this is a new species discovery from the archaeological record. The finds are consistent with species of *Macracanthorhynchus*, of which *M. ingens* is endemic in Arizona. The fact that it is associated with the human-specific pinworm *E. vermicularis* shows that this was indeed an acanthocephalan infection of a human.

The pinworm, *E. vermicularis*, was a universal parasite of Ancestral Puebloans. Enterobiasis is a population-dependent, crowd parasite that varies with population density. Hugot et al. (1999) found that the highest prevalence among Ancestral Puebloans was in stone-walled villages constructed in caves or rock shelters. The villages in Mesa Verde, the Grand Gulch, and Canyon de Chelly represent high-prevalence sites. On an average, 20% of coprolites from these areas are positive for pinworm. Chacoan great houses represented the greatest variation according to Hugot et al. (1999), ranging between 8% and 21%. Reinhard (2008) explored the reasons behind this variation and discovered that when great houses were occupied by smaller populations, infection declined. Smaller Ancestral Pueblo camps had the lowest prevalence range, between 0% and 5%. Five of 22 Antelope Cave human coprolites contained pinworm eggs, resulting in an average prevalence of 22%. This is relatively high and suggests that the people who used Antelope Cave experienced crowding at some point in their annual cycle that promoted crowd diseases such as pinworm.

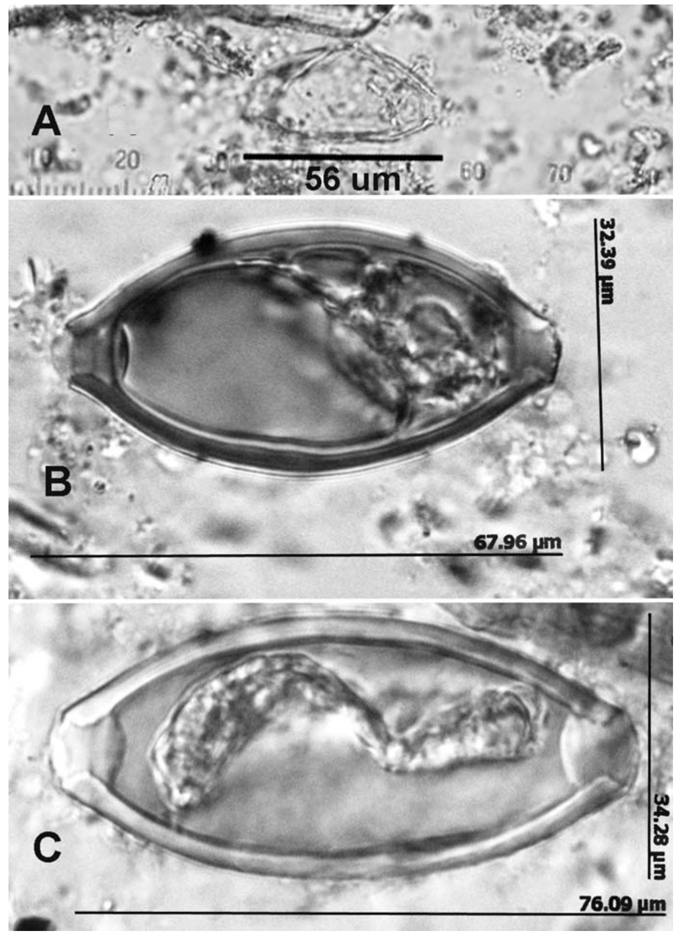

FIGURE 3. Helminth eggs recovered from canid coprolites. A single, poorly preserved capillarid or trichurid egg (**A**) was found in dietary analysis of the coprolite scans. No other eggs of this morphology were found in the parasitological analyses. *Trichuris trichiura* eggs, in comparison, were nearly pristine in preservation. The smallest egg (**A**) showed preservation of one polar plug, but the embryonic mass was compressed by desiccation to one part of the egg. Larger eggs are represented by (**B**). This egg exhibits preservation of both polar plugs and a well-preserved larva.

Other parasite findings from Antelope Cave are of interest. The discovery of a second masticated tick in a human coprolite supports our previous interpretation that ticks were eaten by the inhabitants of Antelope Cave (Fig. 2). We suggest that when the inhabitants found ticks on their bodies, they removed them, crushed them with their teeth, and swallowed them.

The canid coprolites from domestic dogs were positive for *Trichuris vulpis* and perhaps a capillarid (Fig. 3). With respect to the latter possibility, only 1 egg was found, and it was poorly preserved. Repeated efforts to locate more eggs were unsuccessful. Since rabbit and lizard bones were found in the Antelope Cave canid coprolite, this egg could be from a capillarid or trichurid of a prey animal eaten by a dog.

The *T. vulpis* eggs were well preserved but in low numbers. Less C4 that 500 eggs per gram of coprolite were found in 2 of 4 canid coprolites. They ranged in length from 68 to 76 µm and in width from 32 to 34 µm. The preservation of some eggs was pristine, with larvae and polar plugs intact. Even the worst-preserved *T. vulpis* eggs contained some embryonic mass (Fig. 3). The life cycle of *T. vulpis* is monoxenous, without intermediate hosts. The eggs are laid in the intestine, passed in feces, and mature to infective stage in 2–4 wk. The discovery of *T. vulpis* is even more noteworthy because *T. trichiura* has never been recovered from Ancestral Pueblo coprolites (Leles et al., 2010). The find of *T. vulpis* in 2 of 4 coprolites suggests a high number of dogs, since *T. vulpis* is 7 generally found in dogs from high population densities.

By examining coprolites from Antelope Cave, we have expanded our understanding of the totality of Ancestral Pueblo parasitism, both for humans and their companion dogs. It is clear that the diet of Ancestral Puebloans, like other ancient and historic people of the Great Basin, made them susceptible to acanthocephalan infection. The pinworm data suggest that the Virgin River branch of Ancestral Pueblo culture lived in crowded habitations similar to their contemporaries in the Colorado Plateau. Ticks are a unique find at Antelope Cave in human coprolites, as are *T. vulpis* eggs in dog coprolites.

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