

Intestinal Parasitic Infection in the Eastern Roman Empire During the Imperial Period and Late Antiquity

MARISSA L. LEDGER, ERICA ROWAN, FRANCES GALLART MARQUES, JOHN H. SIGMIER, NATAŠA ŠARKIĆ, SAŠA REDŽIĆ, NICHOLAS D. CAHILL, AND PIERS D. MITCHELL

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While there have been numerous studies investigating intestinal parasitic infection in the Roman period, much of this work has been focused in northern Europe, with major gaps in the eastern empire. In order to further elucidate regional patterns in parasitic infection in the Roman empire, we looked for evidence for parasites in sites from Anatolia and the Balkans. Sediment samples from drains as well as coprolites were studied to find evidence for intestinal parasites in the Roman cities of Viminacium (Serbia) and Sardis (Turkey), and results were combined with previous work in these regions. Each sample was tested for preserved helminth (worm) eggs using microscopy and for intestinal protozoa that cause diarrhea, using enzyme-linked immunosorbent assay (ELISA). Our analysis revealed a predominance of species spread by the contamination of food and water by human feces, namely roundworm and whipworm. The identification of these parasites (which are linked to sanitation and hygiene) in Roman cities in Anatolia and the Balkans is contrasted with the range of zoonotic species found elsewhere in the empire. It appears that variations in cooking practices, diet, urbanization, and climate throughout the empire may have contributed to differences in gastrointestinal diseases in different regions.¹

INTRODUCTION

Paleoparasitology, the study of parasites in past populations, is useful for augmenting paleopathological studies on human remains, as it gives us information about a group of infectious diseases that often do not leave evidence on the skeleton. Instead, when individuals are infected with intestinal parasites, their feces contain eggs from the parasite that are very robust. Finding these eggs in archaeological samples known to contain human fecal material can give us evidence for infection in past populations.² Studies in paleoparasitology have found evidence for intestinal helminths (worms) and protozoa (single-celled parasites) in past populations from the Paleolithic to the early modern period.³ As the number of paleoparasitological studies

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²Reinhard et al. 1986.

³Reinhard 1990; P.D. Mitchell 2013, 2015.

increases, it becomes possible to look for regional and temporal variations that were not possible to discern with smaller datasets. The purpose of this study is to contribute evidence for parasitic infection in previously understudied regions of the Roman empire, namely Anatolia and the Balkans, so that we can begin to make regional comparisons. We compare the results from our newly studied sites with what has been found in the rest of the empire to identify patterns. For the Roman period, most work tracing parasitic infection has focused on Britain and France with comparatively little done on the Mediterranean region and the eastern empire.⁴

Intestinal parasites in particular are well suited to such analyses because many parasite infections are chronic and reoccur over an individual's lifetime if they are living in endemic regions. This increases our ability to track them in the archaeological record because infected individuals can excrete eggs in their feces for long periods of time. In excavation of Roman-period settlement areas, the wide-scale presence of drains and latrines provides many opportunities for sampling. In addition, the sediment from the pelvis of skeletons where the intestines decomposed provides opportunities for sampling from excavations of cemeteries as well.

The numerous taxa of intestinal parasites that have been found in archaeological settings⁵ provide evidence that can be used in conjunction with historical sources and archaeological evidence to explore the diet, cooking practices, sanitation and hygiene, migration, and human–animal interactions of past populations.⁶ Parasites are intimately linked with living conditions, diet, food processing techniques, human–animal interactions, and ecology because of their complex life cycles. For example, in the life cycle of beef tapeworm (*Taenia saginata*), eggs released in the feces of an infected human are ingested by cattle, and the immature parasite develops in the animals' tissue. When undercooked beef from an infected animal is eaten by humans, larvae from the meat develop into adult worms in the humans and establish new infections.⁷ Thus, the transmission of this parasite requires

that humans and cattle be interacting on multiple levels: first, that human feces be deposited where the eggs can be ingested by grazing cattle, and, second, that humans rely on these animals as a food source. The same concept applies for other zoonotic parasites that infect humans, such as fish tapeworm and pork tapeworm. The transmission is also dependent on how meat from these animals is prepared, as thoroughly cooking meat will kill larvae. Thus, the presence of beef, pork, or fish tapeworm in archaeological settings allows us to explore diet and cooking practices. By contrast, whipworm (*Trichuris trichiura*) can sustain its life cycle using only humans as a host. The eggs are excreted in the feces of an infected individual and must then develop in the soil for a few weeks; at that point, if they are ingested, the parasite can infect a new individual.⁸ The presence of parasites with life cycles similar to whipworm can allow us to explore the disposal and reuse of fecal material and general sanitation and hygiene.

At its greatest extent, the Roman empire had territory in three continents, including much of Europe, North Africa, and western regions of the Near East. Though there has been much discussion of how culture changed in newly conquered regions of the empire, we also know that the preexisting practices in many areas persisted in the daily life of people living within the borders of the empire.⁹ At the same time, extensive trade networks were established that likely impacted disease transmission, dietary choices, and diffusion of medical practices. These trade networks allowed the introduction of new foodstuffs and the spread of culinary practices—for example, the use of the popular Roman condiment *garum*. For these reasons, we expect that there may be variations in parasite transmission in different regions of the empire. So far, there have been few studies directly comparing disease in different regions of the empire, though these few have shown some variation in health and disease regionally.¹⁰

Currently there is evidence for 10 different taxa of intestinal parasites, helminths, and protozoa, from sites within the Roman empire. These include beef/pork

⁴P.D. Mitchell 2017; Ledger et al. 2018.

⁵Bouchet et al. 2003a; Le Bailly et al. 2003a; Araújo et al. 2015; Seo and Shin 2015; Ledger and Mitchell 2019.

⁶Reinhard 1992; Reinhard et al. 2013; Yeh et al. 2014; Ledger and Mitchell 2019.

⁷Garcia 2016, 435–36.

⁸Garcia 2016, 311.

⁹Woolf 1997; Braund 1998, 22; Huskinson 2009; Garnsey and Saller 2015, 207, 216–22.

¹⁰Gowland and Redfern 2010; Scheidel 2012; Mays et al. 2018.

tapeworm (*Taenia* spp.), *Capillaria* sp. worm, *Entamoeba histolytica* (which can cause dysentery), *Fasciola* sp. liver fluke, fish tapeworm (*Dibothriocephalus* sp. or *Diphyllobothrium* sp.), giardiasis (*Giardia duodenalis*), lancet liver fluke (*Dicrocoelium dendriticum*), pinworm (*Enterobius vermicularis*), roundworm (*Ascaris* sp.), and whipworm (*Trichuris trichiura*).¹¹ This evidence comes from 12 modern countries: Austria, Belgium, Britain, Egypt, France, Germany, Greece, Israel, Italy, the Netherlands, Switzerland, and Turkey. In the Roman period, these regions roughly correspond to the provinces of Pannonia Superior, Belgica, Britannia, Aegyptus, Gallia Aquitania, Gallia Lugdunensis, Gallia Narbonensis, Germania Superior, Raetia, Achaia, Iudaea, Italia, Germania Inferior, Asia, and Lycia et Pamphylia. Recent work by the Ancient Parasites Laboratory at the University of Cambridge has provided the first evidence for parasitic infection in Greece and Turkey during the Roman period. At Ayia Irini on the Aegean island of Kea, eggs from roundworm and whipworm were found in soil from the pelvises of Roman-period burials.¹² In Turkey, at the site of Sagalassos, samples from a communal latrine associated with a bath complex contained the eggs of roundworm and cysts from *Giardia duodenalis*, a protozoan that causes severe diarrhea.¹³ At Ephesus, sediment from a private house latrine contained eggs from whipworm while sediment from a communal latrine contained eggs of roundworm.¹⁴ Here, we combine this evidence with that from additional sites in Anatolia and the Balkans to make interregional and intraregional comparisons, especially with the better-studied provinces in northern Europe, namely those in France and Britain.

In order to find evidence for intestinal parasitic infection in Roman Anatolia and the Balkans, a series of fecal samples and sediment samples that were likely to contain human fecal material were collected from two sites (fig. 1): Sardis in modern-day Turkey (the Roman province of Asia) and Viminacium in Serbia (the Roman province of Moesia). These results are discussed in conjunction with the recently published work by the authors that reports evidence for intestinal parasites from Ephesus, Sagalassos, and Kea, allowing

us to make broader comparisons with other regions for the first time.

MATERIALS

Anatolia

The Roman province of Asia, located in western Turkey, had belonged to Hellenistic kingdoms until 133 BCE. In that year, Attalus III, king of Pergamum, bequeathed his territory in western Anatolia to the Romans.¹⁵ The territory formed the province of Asia with Pergamum as its capital until Ephesus became the capital under the emperor Augustus. In the previously Greek cities in the province of Asia, Hellenistic culture persisted in many ways under Roman control. Greek remained the language spoken by the administration, and Greek identity was reified through civic celebrations, art, athletics, and science.¹⁶ At the same time, material culture in the Greek cities of Anatolia demonstrates the adoption of Roman material culture and practices, such as gladiatorial games and public bathing routines, which shows that both cultures could persist and even meld into a new culture or identity for individuals living in the Roman East.¹⁷ Given the variation in culture and climate in the Greek cities, it is worthwhile assessing whether evidence of parasitic infection in these regions might differ from elsewhere in the empire.

Sardis, one of the newly studied sites, was a major city in the Roman East (see fig. 1). A native Lydian settlement with a long history, it is likely that its mostly indigenous population maintained some aspects of Lydian cultural heritage after the city came under Greek and, later, Roman control.¹⁸ It flourished during the Imperial period, which is when many of its public monuments were built.¹⁹ In late antiquity, Sardis became the capital of the new province of Lydia and flourished in the fourth and fifth centuries CE, which is when its monumental synagogue was constructed.²⁰ It was an urban city with an estimated population in the Late Roman period of about 50,000–100,000 inhabitants.²¹ Like other cities in the eastern Mediterranean, Sardis began to decline in the seventh century CE, with

¹¹ P.D. Mitchell 2017; Williams et al. 2017; Ledger et al. 2018.

¹² Anastasiou et al. 2018.

¹³ Williams et al. 2017.

¹⁴ Ledger et al. 2018.

¹⁵ Magie 1950, 34.

¹⁶ Woolf 1994; van Nijf 2000.

¹⁷ Woolf 1994.

¹⁸ Ratté 2008; Berlin and Kosmin 2019.

¹⁹ Greenwalt 2010.

²⁰ Hanfmann et al. 1983, 160.

²¹ Rautman 2011, 11.



FIG. 1. Location of sites studied in Turkey, Serbia, and Greece. Solid dots are new sites studied here, and open circles are sites we have recently studied; lines indicate borders of Roman provinces ca. 117 CE (drawing by M. Ledger, borders from the Digital Atlas of Roman and Medieval Civilizations [DARMC] Project, Harvard University).

the abandonment of certain public spaces, the subdivision and repurposing of houses, and the misfortune of what appears to have been a catastrophic earthquake.²²

The Balkans

The northeastern frontiers of the Roman empire were located along the Rhine and Danube Rivers in the Balkans. As a result of their location along the boundaries of the empire, many towns in the frontier provinces were built around military forts. Viminacium was the capital of the Roman province of Moesia Superior and was a major legionary fortress and city in Serbia, the Danube region of the Roman empire. Located near the frontier of the empire, it was situated at the junction of major roads important for military movements and trade in the region (see fig. 1). Viminacium was likely established in the first century CE as a military camp with a legion permanently stationed there. Its strategic location and the surrounding rich agricultural land²³ ensured that a thriving city grew around it. Viminacium was a production center for smaller forts in the region; archaeological excavations have

shown that it had workshops to supply goods to other forts and cities in the area.²⁴ The population of the city, estimated to have been about 40,000 at its peak, was quite diverse, with influences from the Celtic Scordisci who lived there in pre-Roman times, the Dacians on the other side of the Danube, and many travelers from elsewhere in the empire.²⁵

Samples Studied

Sediment samples were collected during excavations at Sardis and Viminacium from locations likely to contain human fecal material. At Sardis, sediment was collected from two drains within the city. The first drain was connected to a Late Roman house that had a latrine (trench F55 17.1; fig. 2). The latrine was located off the courtyard of the house, and inhabitants likely had to exit into the courtyard in order to enter the latrine. Based on the building materials and construction, the latrine appears to be a later addition to the house and dates from the late sixth century to the early seventh century CE.²⁶ The latrine was a small

²² Rautman 2011, 24–26.

²³ Nikolić and Bogdanović 2015, 547.

²⁴ Wilkes 2005, 154–59.

²⁵ Golubović and Mrdić 2010.

²⁶ Cahill 2019, 106–7.

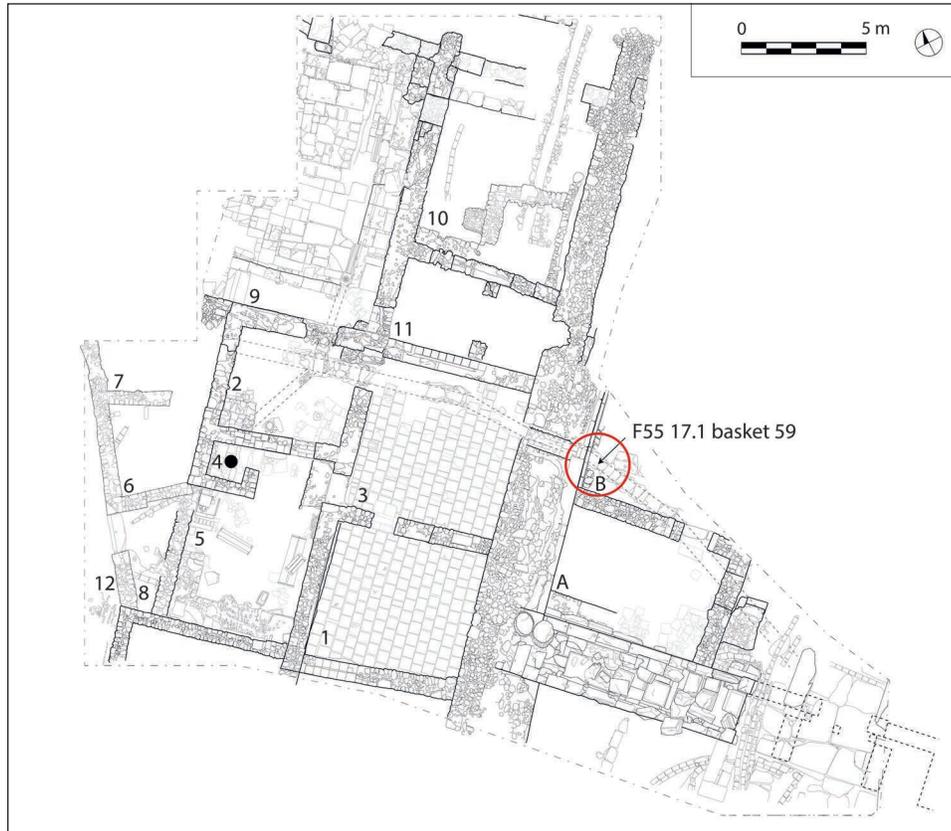


FIG. 2. Plan of a Late Roman house at Sardis (Field 55). Sampling location is marked by the red circle; large drain, marked by dashed lines, connects the sample location to the house latrine, indicated by the black dot (drawing by Archaeological Exploration of Sardis/President and Fellows of Harvard College).

room with an opening across the floor in the rear and a drainage pipe that exited to the north under the adjacent room and connected to a large drain. The large drain ran beneath multiple rooms in the house and also appears to be an addition made in the Late Roman period. Other drains also connected to the large drain,²⁷ and the samples certainly include material from contexts other than this latrine. Therefore, any parasites found in this drain can give evidence for infection in the inhabitants of this house and their guests, as well as in individuals using latrines in other areas of the city connected to this sewer system.

An additional three samples came from drains adjacent to a major avenue on the west side of the city (fig. 3). These drains ran from the south, under the sidewalk of one of the main avenues entering Sardis, where they were sampled, and presumably continued northward and drained into a larger drainage channel

under the paved avenue. This hypothetical main drain, however, has not been identified. The sources of the samples are also uncertain, but they probably originate from houses located on the other side of the modern highway or from shops and workshops fronting the south side of the colonnaded street and facing the Byzantine Shops on the north side of the street. Unfortunately, the modern road prevents us from investigating the question further. Samples were collected from two branches of this sewer that joined together downstream. Similar to the drain near the Late Roman house, this drain dates to the Late Antique period and was probably used beginning in the sixth century CE and into the seventh. Parasite eggs found in the city drain can provide evidence for parasitic infection in the general population of the city.

During the excavations of the city bath complex at the site of Viminacium in 2004, a public communal latrine was identified (fig. 4). Around the perimeter of the room was a deep channel through which water ran to drain feces from the latrine. The (now missing)

²⁷ Cahill 2018, 335–37; Cahill 2019, 105–6.

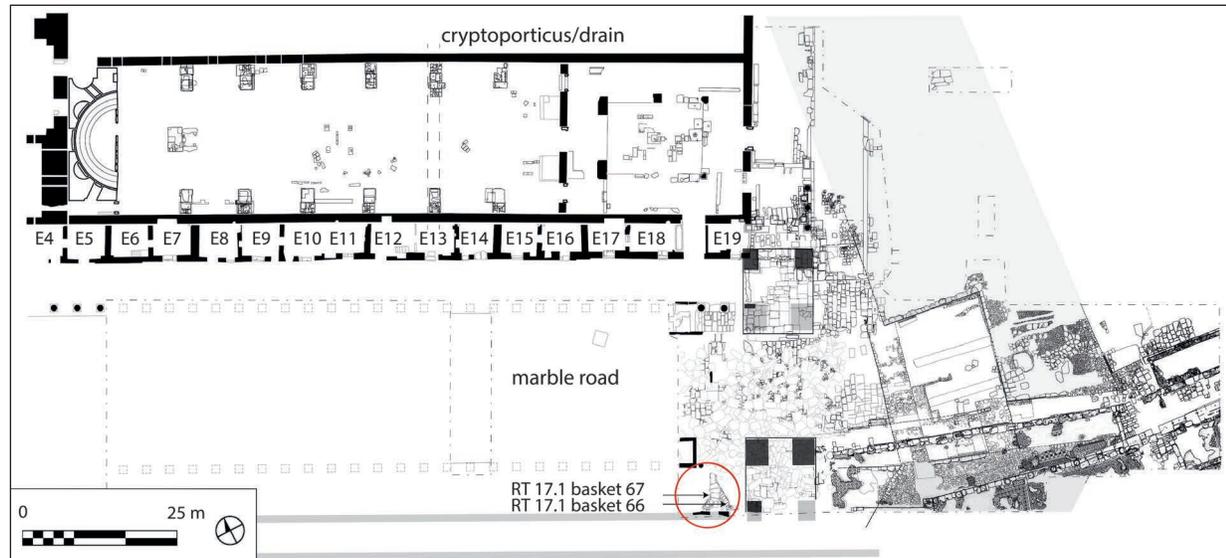


FIG. 3. Plan of synagogue (top), Byzantine Shops (E4–E19), colonnaded avenue, and monumental arch at western entrance to Sardis, showing location of drain samples collected from city drains. Sampling locations are marked by red circle and arrows (drawing by Archaeological Exploration of Sardis/President and Fellows of Harvard College).

toilet seats would have covered this large channel (fig. 5).²⁸ Water would have entered the channel in the southwest corner and left the room at its northwest corner through an opening in the west wall. The flow of water through the room was enabled by the slope of the floor, which is highest in the southwest corner and lowest in the northwest corner. Coprolite deposits were discovered on the floor of the channel near the northeast corner of the room. Coins found on the floor of the room, dating from the second half of the second century CE to the early third, provide a probable date for the use of the latrine and the coprolite deposits found there. A large coprolite found in the drain of the latrine was examined for preserved parasite eggs (see fig. 5, inset). Its surface contour reflects the internal shape of the human colon and rectum. Four separate subsamples were collected from different areas of this coprolite and analyzed.

METHODS

Microscopy

Paleoparasitological analysis of samples from Sardis and Viminacium followed published methods²⁹ and were consistent with the previous analysis undertaken

on samples from Kea, Ephesus, and Sagalassos.³⁰ In brief, 0.2 g subsamples from each sediment sample or coprolite were rehydrated and disaggregated with 0.5% trisodium phosphate. After samples were fully disaggregated into a liquid suspension, they were sieved using a stack of microsieves (with mesh sizes of 300 μm , 160 μm , followed by 20 μm). The material trapped on the 20 μm sieve was collected³¹ and viewed on a digital light microscope at 400x magnification to look for preserved parasite eggs. The entire 0.2 g subsample was viewed (requiring roughly 20 slides per sample), and eggs per gram were calculated by multiplying the number of eggs found in the subsample by five. For the four coprolite subsamples from Viminacium, trisodium phosphate was not sufficient to fully disaggregate the heavily mineralized material, so dilute hydrochloric acid (10%) was added dropwise to break up carbonates until no reaction was visible. The sample was then washed with ultrapure water, centrifuged, the supernatant fluid removed, and this was repeated four times to return the sample to a neutral pH. Use of concentrated hydrochloric acid (36%) for disaggregation has been shown to slightly decrease taxonomic recovery.³²

²⁸ See examples of such latrine architecture in Koloski-Ostrow 2015, 155, 161, 192.

²⁹ Anastasiou and Mitchell 2013.

³⁰ Williams et al. 2017; Ledger et al. 2018.

³¹ Helminth eggs are typically a minimum of 25 μm and maximum of 150 μm in size, so will be trapped on the 20 μm sieve.

³² Dufour and Le Bailly 2013.

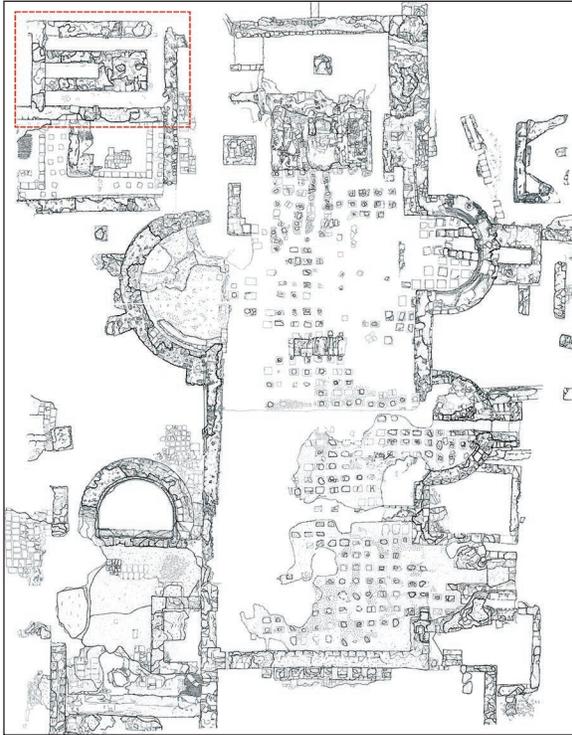


FIG. 4. Plan of bath complex at Viminacium (north at top). Red box indicates the location of the latrine (drawing by N. Šarkić and S. Redžić).

However, dilute acid is less likely to damage parasite eggs; without it, analysis would not have been possible.

ELISA

For the sediment samples from Sardis, we used enzyme-linked immunosorbent assay (ELISA) to test for protozoal parasites that can cause dysentery or severe diarrhea. Direct antigen ELISA tests commercially designed for use with fecal samples (TechLab, Blacksburg, Va.) were employed to detect preserved protozoal antigens in our samples. The three kits used were designed to detect *Entamoeba histolytica*, *Giardia duodenalis*, and *Cryptosporidium* spp. One-gram subsamples were taken from each sample, disaggregated with 0.5% trisodium phosphate, and sieved with the stack of microsieves. For ELISA, the material from below the 20 μm sieve was collected. When individuals are infected with these protozoal parasites, they release cysts in their feces, and these cysts are typically less than 20 μm in size.³³ Thus, the material below the 20 μm sieve



FIG. 5. Latrine at Viminacium. Red arrow indicates location of coprolite found in the latrine; inset shows the coprolite, lgth. 243 mm x width. 146 mm x thickness 60 mm (N. Šarkić and S. Redžić).

should contain the cysts or even preserved antigen in the soil matrix. After collecting the material from the catchment container below the 20 μm sieve, it was centrifuged and the supernatant removed to leave enough sample to be used in each kit, approximately 2.5 ml. An entire column on each ELISA plate was dedicated to each sample, meaning we tested eight replicates for each protozoa. If a positive result was obtained, we repeated the analysis on a new subsample at a later date and only regarded the sample as positive if it was confirmed on both analyses. Each kit uses monoclonal antibodies to detect antigens specific to the protozoa being tested for. The sensitivity and specificity of the tests for modern samples ranges from 96.9% to 100%.³⁴ The manufacturer's procedure was followed for each test. The coprolite samples from Viminacium were not tested with ELISA because it is likely that the dilute acid used in the disaggregation process would impact the preservation of the antigen and affect its interaction with other reagents in the ELISA kits. This could lead to either a false negative or a false positive result.

RESULTS

Sardis

Parasite eggs from roundworm (*Ascaris* sp.) were found in all samples studied from both the city drains and the drain connected to the house latrine at Sardis (fig. 6). In the drain connected to the house latrine,

³³Boone et al. 1999; Tanyuksel and Petri 2003.

³⁴Boone et al. 1999; Gonin and Trudel 2003.



FIG. 6. Roundworm egg from Sardis drain (trench RT 17.1, basket 67). Scale bar is 20 μm (M. Ledger).

roundworm eggs were found in a lower concentration (55 eggs per gram) compared with the city drain, where the concentration ranged from 170 to 305 eggs per gram (table 1). The mean length of the roundworm eggs was 62.3 μm (SD 4.6 μm) and the mean width was 47.8 μm (SD 2.6 μm). All eggs were fertile (unfertilized eggs are a different shape). This fits within standard reported ranges for fertile roundworm eggs in modern humans.³⁵ Recent studies have shown a lack of genetic distinction between the modern species of roundworm that infects humans (*A. lumbricoides*) and the species that infects pigs (*A. suum*) and low host specificity of the two species.³⁶ Thus, we have identified the eggs found to the genus level, as *Ascaris* sp. However, their presence in the drains of a human latrine specifically lends support to their origin in human hosts and thus human infection with roundworm. All three ELISA tests were negative for protozoal parasites in the samples from Sardis.

Viminacium

In the four subsamples studied from the coprolite recovered from the public latrine at Viminacium, eggs

³⁵ Garcia 2016, 1233.

³⁶ *Ascaris lumbricoides* is classically the human-infecting species of roundworm, while *Ascaris suum* is the pig-infecting species. In recent years, modern genetic and epidemiological studies have shown that humans can become infected with *A. suum* and pigs can be infected with *A. lumbricoides*. The eggs from these two species cannot be differentiated based on their morphology. See Betson et al. 2014.

TABLE 1. Egg concentrations (eggs per gram) of roundworm (*Ascaris* sp.) in samples from the Roman drains at Sardis.

Sample	Location	Concentration (eggs/g)
Trench F55 17.1, basket 59	house latrine drain	55
Trench RT 17.1, basket 66, top	city drain	170
Trench RT 17.1, basket 66, bottom	city drain	305
Trench RT 17.1, basket 67	city drain	205

from both roundworm (*Ascaris* sp.) and whipworm (*Trichuris trichiura*) were found (fig. 7). Fertile and infertile roundworm eggs were recovered from the coprolite, and some had preserved mamillated coats.³⁷ The mean length of fertile roundworm eggs was 61.4 μm (SD 4.9 μm) and the mean width was 48.2 μm (SD 4.1 μm) (table 2). The concentration of roundworm eggs in the different subsamples from the same coprolite ranged from 25 to 50 eggs per gram (table 3). This is lower than the concentration found in the drain samples at Sardis, though the Sardis drain samples would have contained fecal material from more than one individual.

The whipworm eggs found were also well preserved, and some still had polar plugs.³⁸ The mean length of eggs with polar plugs was 53.9 μm (SD 3.1 μm) and without polar plugs was 49.5 μm (SD 3.3 μm). The mean width of all eggs was 26.9 μm (SD 1.7 μm) (see table 2). This fits within standard size ranges for modern human whipworm both with and without polar plugs.³⁹ Identification of whipworm eggs to the species level is often done using the size of the eggs.⁴⁰ Human

³⁷ Fertile roundworm eggs are more oval in shape and smaller (average dimensions 45–75 μm long x 35–50 μm wide) compared with infertile roundworm eggs (average dimensions 85–95 μm long x 43–47 μm wide). The mamillated coat is the outer protein layer that causes an appearance of an undulating surface in cross-section; this is often lost in archaeological remains.

³⁸ Polar plugs are the mucoid caps on the ends of the whipworm eggshell where the developing larva will exit the egg.

³⁹ Beer 1976, 48; Garcia 2016, 1233.

⁴⁰ Beer 1976; Confalonieri et al. 1985; Maicher et al. 2017, 1663.

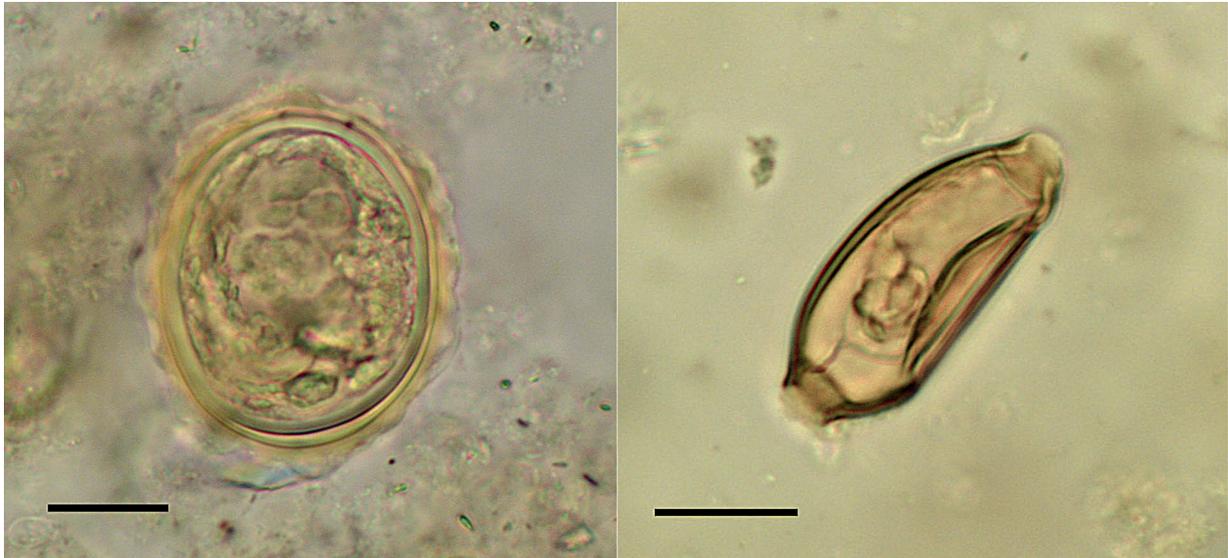


FIG. 7. Helminth eggs found in coprolite from Viminacium: *left*, roundworm egg (subsample 2) with mamillated coat; *right*, whipworm egg (subsample 3); scale bars are 20 μm (M. Ledger).

TABLE 2. Mean egg dimensions and standard deviations (SD) for roundworm (*Ascaris* sp.) and whipworm (*Trichuris trichiura*) from all four subsamples of Viminacium coprolite.

Parasite	Length \pm SD (μm)	Width \pm SD (μm)
<i>Ascaris</i> sp.		
Fertile	61.4 \pm 4.9	48.2 \pm 4.1
Infertile	87.3 \pm 0.0	42.8 \pm 8.5
<i>Trichuris trichiura</i>		
Polar plugs	53.9 \pm 3.1	26.9 \pm 1.7
No polar plugs	49.5 \pm 3.3	

whipworm eggs (*Trichuris trichiura*) and pig whipworm eggs (*Trichuris suis*) are similar in morphology and have overlapping size ranges. The vast majority of eggs found in the Viminacium coprolite fall within the average size range for human whipworm, though this does overlap with the size range of pig whipworm;⁴¹ only five eggs fall outside this human range and within the typical size for pig whipworm exclusively. Based on this and the context of the coprolite, we have identified the eggs as the human-infecting species (*T. trichiura*).

⁴¹ The size ranges used for human and pig whipworm were taken from Beer 1976.

TABLE 3. Egg concentrations (eggs per gram) of roundworm (*Ascaris* sp.) and whipworm (*Trichuris trichiura*) in all four subsamples from Viminacium coprolite.

Subsample	<i>Ascaris</i> sp. Concentration (eggs/g)	<i>T. trichiura</i> Concentration (eggs/g)
1	25	65
2	50	150
3	35	95
4	25	80

The concentration of whipworm eggs in the four different subsamples ranged from 65 to 150 eggs per gram (see table 3). The concentration of whipworm eggs was higher than the concentration of roundworm eggs in each of the subsamples studied. As a single female roundworm produces up to 240,000 eggs per day and a single female whipworm can produce about 7,000 eggs per day, this indicates that the individual the coprolite came from would have had a higher whipworm burden than roundworm burden.⁴²

⁴² Weller and Nutman 2018.

DISCUSSION

Helminth eggs were found in all samples studied. In the coprolite examined from Viminacium in Serbia, eggs of whipworm and roundworm were found. In the drain samples from Sardis, only roundworm eggs were found, in lower concentrations in the drain connected to the house latrine than in the city drain. No other taxa were found in any of the samples studied. When we combine these results with those from our previously published work on samples from Roman-period Sagalassos and Ephesus, two other cities in Anatolia, we can get a better sense of the parasite taxa found in the eastern Mediterranean.

Ephesus and Sagalassos were major urban centers in Anatolia (see fig. 1). Ephesus, made the capital of the province of Asia by Augustus, has been estimated to have had one of the highest populations in the province, and, as a port city, it was certainly an important place for trade, politics, and academia.⁴³ Sagalassos was originally part of the Roman province of Asia but later became part of the neighboring province of Lycia et Pamphylia. It was located along roads leading inland from the Mediterranean and was known for its production of ceramics and crops such as grains and olives.⁴⁴ From a public latrine (second to fifth centuries CE) associated with a bath complex at Sagalassos, eggs of roundworm (10–45 eggs per gram) were found along with antigens from *Giardia duodenalis* cysts, detected with ELISA.⁴⁵ At Ephesus, roundworm eggs were found (17 eggs per gram) in sediment samples from the drain of a sixth-century CE public latrine in the Baths of Scholastikia, while whipworm eggs were found in a third-century CE private house latrine (55 eggs per gram).⁴⁶ Roundworm and whipworm were also the species we found in pelvic sediment from Roman-period burials (146 BCE–330 CE) from the island of Kea in Greece.⁴⁷ When all these sites are taken together, the ubiquity of roundworm and whipworm in the eastern empire and the absence of any zoonotic species become apparent. Though these samples do vary in date and location, they all show a very similar parasite diversity when compared with more northern regions, and they are the first samples that have been

studied in the region of the empire in the eastern Mediterranean and south of the Alps. This prompts us to consider whether similar ecological and sociocultural factors resulted in a consistent parasite profile in this geographical area during this period. As more work is done, it would be beneficial to be able to divide up this large region of the empire for further comparison.

Roundworm, whipworm, and *Giardia duodenalis* (which causes severe diarrhea) are three parasites that are transmitted via the direct fecal-oral route. The life cycle of these parasites can be completed with only one host, in this case humans, though the eggs of roundworm and whipworm do need a period of two to five weeks to mature in the soil before they become infective. After embryonated eggs are ingested by a human, roundworm and whipworm hatch in the intestines. After hatching, roundworm larvae penetrate the intestinal wall and enter the hepatic portal system. They then travel via the circulation to the lungs, to mature for an additional two weeks before ascending the respiratory tract and, after being reswallowed, develop into mature worms in the small intestines. Whipworm larvae mature in the large intestine or colon. Male and female worms then reproduce in the intestines, and eggs are released in the feces of an infected individual.

Most individuals infected with roundworm and whipworm will be asymptomatic or have mild symptoms if the worm burdens are low. However, heavy infections can cause abdominal pain, diarrhea, anemia, and in children can cause stunted growth. High roundworm burdens can cause intestinal obstruction that can be fatal without proper treatment. Roundworms can grow up to 40 cm long, whereas whipworms are smaller at about 3–5 cm. Migration of adult roundworms toward the bile ducts or pancreas can cause acute abdominal conditions such as cholecystitis, cholangitis, or pancreatitis.⁴⁸

As all of our samples came from city drains or latrines that were used by more than one individual, especially in the case of the public latrines, it is difficult to know what proportion of the population may have been infected with these helminths. The egg concentrations, while not extremely high or low, give little indication as to the intensity of infections in the community because it is possible that fecal material in the latrines or sewers was diluted with other material disposed of in these places. Addition of soils, ash, or

⁴³Hanson 2011, 255.

⁴⁴Fuller et al. 2012, 157; Willet and Poblome 2015, 133, 144.

⁴⁵Williams et al. 2017.

⁴⁶Ledger et al. 2018.

⁴⁷Anastasiou et al. 2018.

⁴⁸Jourdan et al. 2018.

sand to cesspits may have helped reduce smells; this was a common practice in the early modern period.⁴⁹ In the Roman period, fecal material in latrines may also have been diluted by kitchen waste. As many latrines were located near kitchens, this was a convenient place to dump cooking water, butchering waste, and other items such as broken ceramic and glass vessels. It is possible that the eggs we found originated from the mixed feces of a few individuals who had high worm burdens and others who had no infection, or that they are from a large number of individuals who had low levels of infection.

Controlling Human Waste in the Roman Empire

Since they are all transmitted by the fecal contamination of food and water, the presence of roundworm, whipworm, and *Giardia duodenalis* in Roman cities in Anatolia and the Balkans gives an indication of sanitation and hygiene levels in these cities. Modern epidemiological work has shown that these parasites are often found in communities with sanitation infrastructure that is not adequate to remove human waste from living areas. Roundworm and whipworm in particular have been found in archaeological samples from settlements across Eurasia from the Neolithic period onward.⁵⁰ The evidence for roundworm and whipworm across the eastern empire suggests that, while public and private latrines were built in cities throughout the Roman empire and were already present in many Greek cities in the east,⁵¹ these facilities did not stop the transmission of gastrointestinal diseases reliant on the fecal-oral route as one might expect. Studies on the distribution of latrines within the city of Pompeii have shown that there is one sanitation feature (a latrine or a downpipe indicating an upper-level latrine) for every two residential properties⁵² in addition to numerous public latrines,⁵³ and in Ephesus, 18 latrines have been identified, with most having multiple seats.⁵⁴ Though the use of toilets has been linked to a decrease in fecal-oral parasite transmission in modern communities, this alone cannot stop transmission; there are various aspects of the design, maintenance, and use of toilets,

as well as other sociocultural practices around hygiene, that may reduce their efficacy.

In Roman cities, the purpose of sewers and latrines was not aimed at stopping the transmission of pathogens, as it is today; the concept of microorganisms was not yet understood at that time. Rather, it is more likely that sewers and latrines were built to remove what was viewed as unclean smells and waste from public view.⁵⁵ The Romans are well known for building large multiseat latrines across the empire (fig. 8 shows an example of a Roman multiseat latrine at Ostia); these are particularly prominent in bath complexes in the Mediterranean. However, it is in Greek cities where some of the early evidence for public multiseat latrines is found, such as at the Aegean islands of Amorgos, Delos, and Thera (fourth to second centuries BCE).⁵⁶ It appears that these sanitation technologies were then adapted by the Romans, who built latrines that could seat even more people at one time. Indeed, most of our evidence for intestinal parasites from Anatolia and the Balkans comes from multiseat latrines such as those at Ephesus, Sagalassos, and Viminacium. (We were not able to collect samples from the multiseat latrines at Sardis.)⁵⁷ Privacy during toilet use was not necessarily expected, and 20 or more seats in one latrine with about 60 cm of space per person was not uncommon.⁵⁸ Evidence from cities such as Pompeii, Herculaneum, Ostia, and Delos have also shown that latrines were located in shops, bars, and possibly even small rooms opening directly off the street that would have been used by passersby when the need arose.⁵⁹ While such facilities served the purpose of giving many people a latrine to use, modern studies have shown that sharing latrines may actually be worse for the spread of disease than a lack of latrines.⁶⁰ This is likely because of the increased exposure to the fecal material of other people.⁶¹

Private latrines have been found commonly in major urban centers in the Roman Mediterranean, but these also had their pitfalls. Many private latrines were

⁴⁹ Petznek et al. 2011, 105.

⁵⁰ Anastasiou 2015.

⁵¹ Gräzer et al. 2011, 29–39.

⁵² Trusler 2017.

⁵³ Koloski-Ostrow 2015, 5–10.

⁵⁴ Jansen 2006.

⁵⁵ Koloski-Ostrow 2015, 66.

⁵⁶ Antoniou and Angelakis 2015, 53, 62–64. See also Antoniou 2007, 156; 2010, 79.

⁵⁷ Yegül 1986, 22.

⁵⁸ Petznek et al. 2011, 99–106.

⁵⁹ Hobson 2009, 55–60; Pérez et al. 2011, 126–28; Rowan 2017.

⁶⁰ Oswald et al. 2017.

⁶¹ Emch 1999.



FIG. 8. Forum latrine, Ostia, Italy; example of a Roman multi-seat latrine (P. Mitchell; courtesy Archivo Fotografico del Parco Archeologico di Ostia Antica).

located in or adjacent to kitchens.⁶² The design of private latrines varies across the empire. In general, many had one or two seats, and most were not connected to main sewers but had cesspits to collect excrement.⁶³ For those that were connected to sewers, most did not have running water and would have needed to be flushed by pouring water down the latrine. An open cesspit, or a poorly drained latrine connecting to public sewers, would have posed a risk for microscopic parasite eggs to be transferred from the latrine into the living spaces. In particular, for latrines located near kitchens, flies may have easily transmitted eggs from the latrine into the food preparation areas or the food itself, as the parasite eggs could adhere to the flies' legs when they landed in the latrine and then be carried to other surfaces. The eggs of roundworms in particular are sticky and are known to be transmitted by flies to human living spaces, where they may then be ingested and cause infection.⁶⁴

There are, of course, many other places where one could come into contact with fecal material in a Roman city aside from the latrines. Texts painted and carved on walls and monuments in Roman cities in the Mediterranean give us some indication that not everyone in these cities used latrines. The numerous warnings against defecating on graves, beside houses, and in other public places⁶⁵ indicate that this happened frequently enough to be an annoyance. There is also evidence for the use of chamber pots in Roman cities, especially by the elite, and these would have been regu-

larly emptied by slaves either into latrines within the house or somewhere acceptable outside the home.⁶⁶ It has even been suggested that elites might have preferred to use chamber pots at home and that latrines located on the main floor of residences were primarily for slaves or people working in the house.⁶⁷ For residences with no latrines, fecal material would have been collected and dumped into latrines, open sewers, the street, or on dung heaps.⁶⁸ Cesspits would have required regular emptying and sewers regular cleaning, which would have been a job for slaves or *stercorarii* (individuals whose job it was to empty cesspits and collect manure). A similar job seems to have existed in Hellenistic cities⁶⁹ and likely persisted into the Roman period in Anatolia. The number of individuals handling fecal material and disposing of it outside urban centers may have had a considerable impact on the transmission of intestinal parasites, and certainly these individuals themselves would be at high risk for infection.

Finally, the reuse of human fecal material as fertilizer is mentioned in Roman texts on agriculture. Varro cites the work of Cassius, who stated that human fecal material was second only to pigeon dung as a fertilizer (Varro, *Rust.* 1.38.1–3). Similarly, Columella in his texts on agriculture discusses three types of manure, that from birds, humans, and cattle; he ranks human excrement after that from birds if it is mixed with other refuse (Columella, *Rust.* 2.14.1–8). From the cesspits of private latrines, fecal material may have been reused on gardens of the same home, or it could have been collected by *stercorarii* and sold as manure.⁷⁰ If it was in fact reused in this way, especially in private gardens where vegetables and fruits were grown, parasite eggs from infected individuals could easily be ingested on unwashed fruits and vegetables, thus transmitting roundworm, whipworm, and dysentery in these communities.

Regional Variation

Having established potential reasons for the presence of roundworm and whipworm in these sites in the eastern region of the empire, we can now turn to how

⁶² Pérez et al. 2011, 113–18.

⁶³ Hoss et al. 2011, 51–55.

⁶⁴ Hall 1982.

⁶⁵ Wilson 2000, 310–11.

⁶⁶ Hobson 2009, 69.

⁶⁷ Trusler and Hobson 2017.

⁶⁸ Taylor 2015, 75.

⁶⁹ Owens 1983.

⁷⁰ Flohr and Wilson 2011, 147–48.

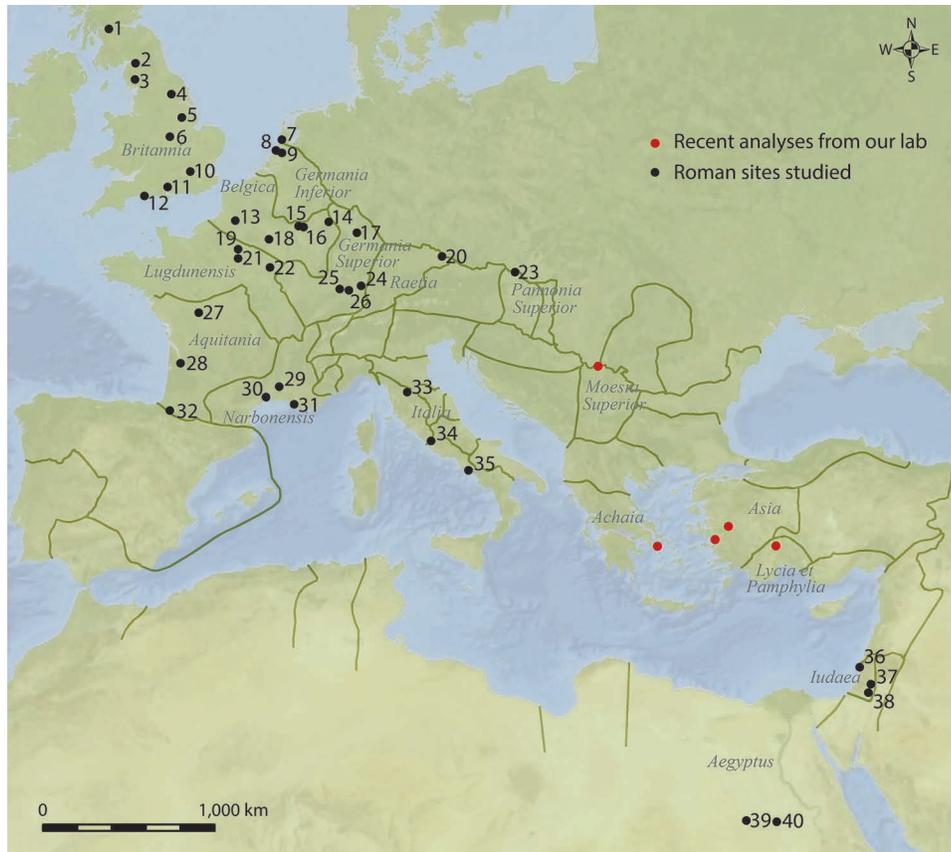


FIG. 9. Map of the Roman empire indicating sites where evidence for intestinal parasites has been found. Red dots indicate sites recently analyzed by our lab (see fig. 1); lines indicate borders of Roman provinces ca. 117 CE. 1, Bearsden; 2, Carlisle; 3, Ambleside; 4, Church Street Sewer; 5, Lincoln Waterside; 6, Leicester; 7, Utigeest; 8, Valkenburg Army Camp; 9, Alphen on the Rhine; 10, London; 11, Owslebury; 12, Poundbury; 13, Amiens; 14, Belginum; 15, Mageroy; 16, Arlon; 17, Ladenburg; 18, Reims; 19, Bobigny Hospital; 20, Künzing; 21, Lisses; 22, Troyes; 23, Carnuntum; 24, Eschenz; 25, Augst; 26, Vindonissa; 27, Jaunay-Clan; 28, Bordeaux; 29, Le Gramiere; 30, Lattes; 31, Marseille; 32, Mikelauen-Zilo; 33, Uffizi Gallery Burials; 34, Roma; 35, Pompeii; 36, Caesaria; 37, Qumran; 38, Nahal-Mishmar Valley; 39, Dakhleh Oasis; 40, El-Deir (drawing by M. Ledger, borders from the Digital Atlas of Roman and Medieval Civilizations [DARMC] Project, Harvard University).

this taxonomic diversity compares with other regions of the empire. The five sites studied in Greece, Anatolia, and the Balkans do vary in size and location, and the samples come from varying dates. As such, they are giving us a broad overview of patterns of parasite diversity in this area of the empire. Previous paleoparasitological work done on other Roman sites also covers a broad range of site types (e.g., military, urban, rural) and uses a range of sample types (table 4). Dates similarly span the entirety of the Roman period. As more data become available, site locations and characteristics, sample types, and dates can be more tightly controlled, allowing for more precise comparisons.

Most paleoparasitological studies performed at Roman-period sites have taken place in northern Europe (fig. 9; see table 4).⁷¹ From the Roman provinces of Belgica, Germania Inferior, and Germania Superior—which cover parts of modern-day Belgium, Germany, the Netherlands, and Switzerland—beef/pork tapeworm, *Capillaria* worm, *Entamoeba histolytica*, *Fasciola* liver fluke, lancet liver fluke, roundworm, and whipworm have all been found.⁷² From the province

⁷¹ Anastasiou 2015; P.D. Mitchell 2017.

⁷² Kuijper and Turner 1992; Jauch 1997; Dittmar et al. 2002; Defgnée et al. 2008; Mowlavi et al. 2014; Le Bailly and Bouchet 2015.

TABLE 4. Details of previous paleoparasitological studies that have found intestinal parasites in Roman-period samples, listed first by country and then chronological order. Table includes the modern-day country where the site is located, the number marked on fig. 9, the date of the samples studied, the parasite taxa found as reported by the authors, the type of sample studied, and the reference.

Country	Map Label	Site	Date	Parasites	Sample Type	Reference
Austria	23	Carnuntum	101–300 CE	<i>Ascaris lumbricoides</i> <i>Taenia</i> sp. <i>Trichuris trichiura</i>	sewer and latrine	Aspöck et al. 2011; Petznek 2018
Belgium	16	Arlon	1–300 CE	<i>Ascaris</i> sp. <i>Trichuris</i> sp.	vats or pits	Defgnée et al. 2008; Gourevitch et al. 2011
Britain	15	Mageroy	Roman	<i>Entamoeba histolytica</i>	latrine	Le Bailly and Bouchet 2015
	6	Leicester	1–200 CE	<i>Ascaris</i> sp. <i>Fasciola</i> sp. <i>Trichuris</i> sp.	cesspit	Boyer 1999
	10	London, Hibernia Wharf	1–200 CE	<i>Ascaris lumbricoides</i> <i>Diphyllobothrium latum</i> <i>Taenia</i> sp. <i>Trichuris trichiura</i>	well	Rouffignac 1985
	2	Carlisle	1–300 CE	<i>Ascaris lumbricoides</i> <i>Fasciola</i> sp. <i>Trichuris trichiura</i>	occupation sediment	Jones and Hutchison 1991
	3	Ambleside	1–400 CE	<i>Ascaris</i> sp. <i>Trichuris trichiura</i>	pit	Jones 1985
	4	Church Street Sewer, York	1–500 CE	<i>Ascaris</i> sp. <i>Trichuris</i> sp.	sewer	Wilson and Rackham 1976
	10	London, 15–35 Cophthall Ave.	101–400 CE	<i>Dicrocoelium dendriticum</i> <i>Trichuris trichiura</i>	occupation sediment	de Moulins 1990
	1	Bearsden	142–158 CE	<i>Ascaris</i> sp. <i>Trichuris</i> sp.	sewer	Knights et al. 1983
	5	Lincoln, Waterside NW	301–400 CE	<i>Trichuris</i> sp.	occupation sediment	Carrott et al. 1995
	11	Owslebury, Winchester	Roman	<i>Ascaris</i> sp. <i>Dicrocoelium</i> sp. <i>Trichuris/Capillaria</i> sp.	pit	Pike 1968
	12	Poundbury, Dorset	Roman	<i>Ascaris lumbricoides</i> <i>Trichuris trichiura</i>	coffin sediment	Jones 1987
Egypt	39	Dakhleh Oasis	395 BCE–30 CE	<i>Enterobius vermicularis</i>	mummy	Horne 2002
	40	El-Deir	301–500 CE	<i>Taenia</i> sp.	mummy	Le Bailly et al. 2010

France	19	Bobigny Hospital	201 BCE–100 CE	<i>Ascaris lumbricoides</i> <i>Trichuris trichiura</i>	pelvic soil	Rousset et al. 1996
	31	Marseille	14 BCE–27 CE	<i>Diphyllobothrium</i> sp. <i>Fasciola</i> sp. <i>Trichuris</i> sp.	occupation sediment	Harter-Lailheugue 2006
	30	Lattes	1–200 CE	<i>Entamoeba histolytica</i>	cesspit	Le Bailly and Bouchet 2006
	28	Bordeaux	40–51 CE	<i>Ascaris</i> sp. <i>Taenia</i> sp. <i>Trichuris</i> sp.	sewer or latrine	Sireix 2008
	18	Reims	101–200 CE	<i>Ascaris</i> sp. <i>Fasciola</i> sp. <i>Trichuris</i> sp.	pits, wells, occupation sediment	Bouchet et al. 2001; Le Bailly et al. 2003a
	27	Jaunay-Clan	201–300 CE	<i>Trichuris trichiura</i>	coffin sediment	Dufour et al. 2016
	29	La Gramiere, Castillon du Gard	201–300 CE	<i>Entamoeba histolytica</i>	cesspit	Gonçalves et al. 2004
	13	Amiens	201–400 CE	<i>Capillaria hepatica</i>	cyst with skeletal remains	Mowlavi et al. 2014
	21	Lisses	Roman	<i>Entamoeba histolytica</i>	pit	Le Bailly and Bouchet 2015
	32	Mikelauen-Zilo	Roman	<i>Dicrocoelium</i> sp. <i>Diphyllobothrium</i> sp.	occupation sediment	Le Bailly and Bouchet 2010, 2013
	18	Reims, Rue Carnot	Roman	<i>Dicrocoelium</i> sp.	cesspit	Le Bailly and Bouchet 2010
	22	Troyes, Place de la Liberation	Roman	<i>Dicrocoelium</i> sp. <i>Entamoeba histolytica</i>	cesspit	Le Bailly and Bouchet 2010, 2015
Germany	17	Ladenburg	1–200 CE	<i>Ascaris</i> sp. <i>Trichuris</i> sp.	latrine	Goppelsröder and Sommer 1996
	14	Belginum	101–300 CE	<i>Ascaris</i> sp. <i>Capillaria</i> sp. <i>Taenia</i> sp.	pit	Dittmar et al. 2002
	20	Künzing	140–250 CE	<i>Trichuris trichiura</i>	pit	Specht 1963
Israel	37	Qumran	100 BCE–68 CE	<i>Ascaris lumbricoides</i> <i>Dicrocoelium</i> sp. <i>Enterobius vermicularis</i> <i>Taenia</i> sp. <i>Trichuris trichiura</i>	latrine	Harter et al. 2004; Zias et al. 2006
	38	Nahal-Mishmar Valley	160 CE	<i>Trichuris trichiura</i>	coprolites	Witenberg 1961
	36	Caesaria	Roman	<i>Ascaris lumbricoides</i> <i>Diphyllobothrium</i> sp.	latrine	Harter 2003

Country	Map Label	Site	Date	Parasites	Sample Type	Reference
Italy	35	Pompeii	100 BCE–79 CE	<i>Trichuris trichiura</i>	sewer	Heirbaut et al. 2011
	33	Uffizi Gallery Burials	301–500 CE	<i>Ascaris</i> sp.	pelvic soil	Roche et al. 2019
	34	Roma	Roman	<i>Entamoeba histolytica</i>	–	Le Bailly and Bouchet 2015
Netherlands	7	Uffigeest	101 BCE–300 CE	<i>Trichuris</i> sp.	well	van Geel et al. 2003
	9	Alphen on the Rhine	1–100 CE	<i>Ascaris</i> sp. <i>Taenia/Echinococcus</i> sp. <i>Trichuris</i> sp.	latrine	Kuijper and Turner 1992
	8	Valkenburg Army Camp	1–100 CE	<i>Ascaris lumbricoides</i> <i>Trichuris trichiura</i>	occupation sediment	Jansen and Over 1966
Switzerland	25	Augst	1–100 CE	<i>Ascaris</i> sp. <i>Trichuris</i> sp.	latrine	Hänggi et al. 1989; Hufschmid and Sütterlin 1992
	24	Eschenz	1–200 CE	<i>Ascaris lumbricoides</i> <i>Trichuris trichiura</i>	latrine	Jauch 1997
	26	Vindonissa	30–45 CE	Ascarididae <i>Trichuris</i> sp.	latrines	Le Bailly et al. 2003b

of Britannia, beef/pork tapeworm, *Fasciola* liver fluke, fish tapeworm, lancet liver fluke, roundworm, and whipworm have been found.⁷³ At one site, Carnuntum, in the province of Pannonia Superior (Austria), roundworm, whipworm, and beef/pork tapeworm were found.⁷⁴ At Künzing, in the province of Raetia, whipworm was found.⁷⁵ From the provinces of Aquitania and Lugdunensis, which cover most of modern-day France, beef/pork tapeworm, *Entamoeba histolytica*, fish tapeworm, lancet liver fluke, roundworm, and whipworm have been found.⁷⁶

The province of Narbonensis, in southern France, can be considered part of the Mediterranean basin. Here, there is evidence for *Entamoeba* dysentery, *Fasciola* liver fluke, fish tapeworm, and whipworm.⁷⁷ In the central and eastern Mediterranean region, Roman sites have been studied from Israel and Italy, and now Greece, Turkey, and Serbia. A species diversity similar to northern Europe has been found in Israel (province of Iudaea), where evidence exists for beef/pork tapeworm, fish tapeworm, lancet liver fluke, pinworm, roundworm, and whipworm.⁷⁸ In Egypt, mummified remains have given evidence for pinworm and beef/pork tapeworm.⁷⁹ However, in Greece, Italy, and Turkey, so far there is only evidence for *Giardia duodenalis*, roundworm, and whipworm,⁸⁰ the same taxa found at the sites analyzed in this study. One commonality we see across the empire is that fecal-oral parasites, especially roundworm and whipworm, were present in all study regions. However, in many provinces they are found alongside other zoonotic species. There is no evidence in Greece, Italy, and Turkey for the beef/pork tapeworms, fish tapeworm, and liver flukes that have been found in other regions (table 5).

The absence in Greece, Italy, and Turkey of certain taxa of intestinal parasites is difficult to interpret because we do not know the degree to which certain eggs may have been lost due to decomposition over time.

⁷³ Rouffignac 1985; de Moulins 1990; Jones and Hutchison 1991.

⁷⁴ Aspöck et al. 2011; Petznek 2018.

⁷⁵ Specht 1963.

⁷⁶ Rousset et al. 1996; Le Bailly and Bouchet 2006, 2010, 2013, 2015; Sireix 2008; Dufour et al. 2016.

⁷⁷ Harter-Lailheugue 2006; Le Bailly and Bouchet 2006.

⁷⁸ Harter 2003; Zias et al. 2006.

⁷⁹ Horne 2002; Le Bailly et al. 2010.

⁸⁰ Heirbaut et al. 2011; Le Bailly and Bouchet 2015; Williams et al. 2017; Anastasiou et al. 2018; Ledger et al. 2018.

TABLE 5. Overview of types of intestinal parasites found in previously published and currently studied sites in provinces in Asia (Sardis, Ephesus), Lycia et Pamphylia (Sagalassos), and Moesia Superior (Viminacium) compared with evidence from other Roman provinces.

Province	Parasites Typically Transmitted by the Direct Fecal-Oral Route				Zoonotic Parasites			
	Whip-worm	Round-worm	Protozoa (dysentery)	Pinworm	<i>Capillaria</i> sp.	Beef/pork tapeworm	Fish tapeworm	Liver flukes
In Provinces Bordering the Mediterranean								
Aegyptus	–	–	–	✓	–	✓	–	–
Iudaea	✓	✓	–	✓	–	✓	✓	✓
Asia	✓	✓	–	–	–	–	–	–
Lycia et Pamphylia	–	✓	✓	–	–	–	–	–
Moesia Superior	✓	✓	–	–	–	–	–	–
Achaia	✓	✓	–	–	–	–	–	–
Italia	✓	–	✓	–	–	–	–	–
Narbonensis	✓	–	✓	–	–	–	✓	✓
In Provinces Outside the Mediterranean Region								
Aquitania	✓	✓	–	–	–	✓	✓	✓
Lugdunensis	✓	✓	✓	–	–	–	–	✓
Pannonia Superior	✓	✓	–	–	–	✓	–	–
Raetia	✓	–	–	–	–	–	–	–
Germania Superior	✓	✓	–	–	–	–	–	–
Germania Inferior	✓	✓	–	–	–	✓	–	–
Belgica	✓	✓	✓	–	✓	✓	–	✓
Britannia	✓	✓	–	–	–	✓	✓	✓

The samples that have been studied from sites in other regions are a mix of latrine or cesspit sediment, drain sediment, occupation layer sediment, pelvic soil, and Egyptian mummies; thus, they are broadly similar to what we have studied in Turkey and Serbia except for the mummified remains (see table 4 for information on sample types from other studies). It is possible that some parasite taxa were not represented in the samples from Turkey and Serbia because they were present in lower concentrations that were not picked up during analysis or because the eggs did not preserve as well as those of roundworm and whipworm. However, as the eggs of many of these species are just as robust as whipworm and roundworm (which we did find), we suspect that zoonotic parasites were just not as common in the eastern part of the empire. Infection with beef/pork tapeworms, fish tapeworm, lancet liver

fluke, *Fasciola* liver fluke, and *Capillaria* generally require interaction with infected animals, either by ingestion of poorly cooked liver, meat, or fish, or by ingestion of aquatic plants, from water contaminated with animal feces, that have larvae encysted on them, as in the case of *Fasciola*.⁸¹

Anatolia was a heavily urbanized region of the Roman empire.⁸² The degree of urbanization and population density could have contributed to differential transmission of certain parasites. The sites we have studied in the provinces of Asia and Lycia et Pamphylia are all urban cities, where individuals might have had less interaction with animals than they would in

⁸¹ Ledger and Mitchell 2019.

⁸² Hanson 2011.

rural settings. However, as the number of individuals in a city increased with urbanization, the removal of human waste from the city would have become more of a challenge. This could have increased the transmission and prevalence of fecal-oral parasites if the removal processes were inadequate. While Viminacium was not located in the heavily urbanized province of Asia, we know it was a large military fortress and one of the most densely populated centers in the province of Moesia Superior.

Absence of Zoonotic Parasites in the Eastern Empire

There are numerous potential explanations for the absence of zoonotic parasites in the sites in the eastern empire. Perhaps meat was not widely eaten by all inhabitants of these towns (especially those of the lower classes), or perhaps meat and fish were generally well cooked. Where animals were herded, they had other longer-term uses that precluded butchering purely for meat;⁸³ for example, goats could be used for milk and cheese, sheep for their wool (Columella, *Rust.* 7.1.2), and cattle as work animals (Columella, *Rust.* 6 *praef.* 1–3). As such, they would only be slaughtered in old age or when they were sick (Columella, *Rust.* 7.7.2). Cereals, legumes, olive oil, and wine have been suggested to have been core foods in the Roman diet in the Mediterranean,⁸⁴ and these do not pose a risk for transmission of foodborne tapeworms. Currently, there has been one stable isotope study done on human remains from Ephesus.⁸⁵ This study has shown that crops such as wheat and barley were likely staple foods; and looking at the nitrogen isotope ratios, the low $\delta^{15}\text{N}$ values compared with those from other regions such as Roman Italy are suggested to be a result of frequent legume consumption along with some animal protein.⁸⁶ Stable isotope studies on human remains from Sagalassos similarly had low $\delta^{15}\text{N}$ values in comparison with elsewhere in the Mediterranean, possibly suggest-

ing somewhat lower reliance on marine resources than in Italy, in conjunction with reliance on domesticated animals such as pigs, sheep, goats, and cattle.⁸⁷ These studies exemplify a diet based on cereals with additions of different types of meat and comparatively low reliance on marine resources, though the frequency at which meat was consumed cannot be determined.

Cultural influences in the Greek East may have further impacted diet in these cities. A regional exploration of diet in the Roman empire based on zooarchaeological remains has shown that there were distinct variations in diet in the different provinces. In particular, Roman cities in the Greek East seem to be heavily influenced by and consistent with Hellenistic patterns that mainly relied on goat and sheep with the minor addition of beef and pork (but not at the higher levels seen in Roman Italy).⁸⁸ Zooarchaeological remains from Sagalassos generally follow this pattern with sheep/goat consistently found in high proportions compared with other domesticates, though cattle become distinctly more important in the Late Imperial period.⁸⁹ We may expect lower levels of beef and pork tapeworm infection if sheep and goat were more often used for meat. Pork was eaten at Sagalassos throughout the Imperial period and late antiquity; however, pig remains are not found in higher proportions than sheep/goat or cattle in any time period, and they peak at 25% in the Late Imperial period.⁹⁰ In zooarchaeological assemblages from Viminacium, cattle were dominant, accounting for about half the assemblage, followed by pig and then sheep/goat.⁹¹ The location where herded animals were kept would have also influenced the risk of ingesting parasite eggs from human feces; this is especially the case for cattle and pigs, which need to ingest *Taenia* sp. eggs from human feces to maintain the life cycle of beef and pork tapeworm. Free-ranging pigs may have been less likely to ingest human fecal material than those raised in sties within the city or herded in areas with fertilized crops. Studies using stable isotopes, linear enamel hypoplasias (LEHs), and dental microwear of pig teeth at Sagalassos have suggested that at least some pigs were free-ranging for most of their life and kept in sties just before slaughtering.⁹²

⁸³ Chandezon 2015, 136.

⁸⁴ Garnsey 1999, 13–17; S. Mitchell 2015.

⁸⁵ Lösch et al. 2014.

⁸⁶ Lösch et al. 2014, 9. Other isotope studies done in Croatia, Italy, and Greece also show a general reliance on C_3 plants (these include wheat and barley, as opposed to C_4 plants, which include sorghum and millet) with minor additions of fish and terrestrial animals for protein, though there is some variation at different sites. See Prowse et al. 2005; Lightfoot et al. 2012; Dotsika and Michael 2018; McConnan Borstad et al. 2018; O'Connell et al. 2019.

⁸⁷ Fuller et al. 2012.

⁸⁸ King 1999.

⁸⁹ Fuller et al. 2012.

⁹⁰ Fuller et al. 2012, 160.

⁹¹ Vuković-Bogdanović 2016.

⁹² Vanpoucke et al. 2009; Frémondeau et al. 2017.

In northern Europe, dental microwear analysis from Elms Farm in Roman Britain suggests that pigs were stall fed, though sites studied in Gaul suggest pigs there were free-ranging.⁹³

Longstanding social and religious practices and expectations in the Greek East likely influenced how often meat was consumed. Roman writers, noting the otherness of nomadic tribes with whom the Romans came into contact during the expansion of the empire,⁹⁴ describe their reliance on animal meat and milk as though this reliance on meat was unusual for the typical urban Roman elite. From Tacitus, we learn that German tribes were known for eating meat, in particular fresh game (Tac., *Germ.* 23). However, caution is needed when interpreting the opinions of Roman elite writers who might never have traveled to these regions and were heavily influenced by cultural biases; they may give us a view of the expectations for elite Roman diet rather than a holistic view of the variations in diet across social classes and regions of the empire.⁹⁵ It has been suggested, for example, that meat eating was more common in northwestern Europe than in the Mediterranean.⁹⁶ A wide-scale comparison of isotopic data from different regions of the empire could provide evidence for variations in levels of meat consumption.

We should also consider that access to meat could have been influenced by social class, occupation, age, sex, and personal preference. For example, the Roman military is often described as relying on meat to form part of their diet.⁹⁷ The diet of the military likely differed from civilians living in the same town. Despite this, we have not found any evidence for tapeworms at Viminacium, which had a legion permanently stationed there.

The preparation of meat is also very important regarding the transmission of tapeworms. Eating raw meat (aside from salted products) was considered to be the dietary practice of barbarians;⁹⁸ Tacitus clearly disapproves of the Fenni in northern Europe who ate the half-raw flesh of animals.⁹⁹ Most of the numerous recipes for meat dishes, or sauces to be put on meat,

found in Apicius' *De re coquinaria* call for the meat to be boiled or roasted. These rather elaborate recipes reflect dishes that would have been prepared by cooks working in the homes of wealthier Romans, and it is not known whether such dishes were dispersed across the empire. One recipe for smoked pork calls for first smoking the pork and then boiling it in salt water before eating it (Apicius, *De re coquinaria* 8.7.380). Ancient descriptions of how to prepare salted pork suggest that it was eaten this way to some extent (Cato, *Agr.* 162.1–3).¹⁰⁰ Preparing meat by salting can kill parasite larvae; however, very high levels of salt for extended periods of time are required to kill larvae effectively.¹⁰¹ Similarly, thorough cooking of meat can kill any parasite larvae and stop the transmission of parasites like beef and pork tapeworm, but even if meat was generally cooked, there might have been low levels of tapeworm transmission as a result of meat not being thoroughly cooked every time it was prepared. Thus, cases of tapeworm could be more common in certain regions or in groups of individuals that ate meat more often.

All the sites studied in Anatolia and Serbia are large cities or military sites. If individuals living in these urban centers were infected with beef or pork tapeworms, we could find these eggs in town drains and latrines. The absence of robust beef and pork tapeworm eggs suggests that infections were not common in the region, perhaps due to low levels of meat consumption, or the general scarcity of beef and pork in the diet, or the tendency to raise live animals at a distance from the city. It is also possible that when beef and pork were consumed, local cooking practices, in which the meat was thoroughly cooked, stopped the transmission of these parasites.

The popularity of salted fish or fish sauce (*garum* and *liquamen*) in the Roman empire is well known and has been suggested as a potential cause of fish tapeworm infections where this parasite has been found in northern Europe.¹⁰² The Mediterranean region is known for its production of salted fish and fish sauces, and the salting process for preserving fish is thought to have originated in the east and spread to the rest of the Mediterranean and later throughout the Roman

⁹³ Vanpoucke et al. 2007; Wilkie et al. 2007, 252.

⁹⁴ Garnsey 1999, 65–68.

⁹⁵ Prowse et al. 2004, 260; Killgrove and Tykot 2013.

⁹⁶ Garnsey 1999, 65; Chandezon 2015, 138.

⁹⁷ Davies 1971, 126; Garnsey 1999, 17; Chandezon 2015, 144.

⁹⁸ Chandezon 2015, 142.

⁹⁹ Garnsey 1999, 68.

¹⁰⁰ Chandezon 2015, 140.

¹⁰¹ Khalil 1969; Adams et al. 1997.

¹⁰² P.D. Mitchell 2017.

empire.¹⁰³ The species of fish tapeworm found in Europe use freshwater fish and fish that spend a portion of their life in freshwater as an intermediate host.¹⁰⁴ It is unclear if the salting process used to make *garum* was adequate to kill fish tapeworm larvae found in fish. However, it has been suggested that most salted fish products were made from marine fish,¹⁰⁵ which cannot act as an intermediate host to *Diphyllobothrium* sp. in this region of the world. Rather, consumption of local freshwater fish that were inadequately cooked may be responsible for cases of fish tapeworm in the Roman empire.

Finally, climate in the Mediterranean likely has an impact on parasite diversity. Though this can be difficult to disentangle from the sociocultural impacts on parasite transmission, we have to consider that it also played a role. The Mediterranean region is generally warmer and drier than northern Europe.¹⁰⁶ The warmer climate would increase the rate at which meat and fish spoiled if not cooked or preserved with extensive salting or smoking, and it is possible that heavy salting of meats in the more southerly regions stopped the transmission of some zoonotic species. The overall drier climate at certain times of the year in eastern Mediterranean regions, as compared with northern Europe, could affect transmission of certain parasites that need aquatic conditions to survive, such as fish tapeworm and *Fasciola* liver fluke. Paleoclimate reconstructions have indicated that there was an almost century-long drought in Anatolia from ca. 350 to 470 CE.¹⁰⁷ This drought would have affected the length of growing seasons and the crops that could have been supported. Crop failures may have increased the perceived need for and use of fertilizer, some of which could have contained human feces and thus spread fecal-oral pathogens. Paleoparasitological studies from all time periods have shown that the ecology of sites is an important determinant of taxonomic diversity. The highest diversity of parasites in archaeological sites in Europe is often found in lakeside regions.¹⁰⁸ This is likely a result of an increase in species that can complete their life cycles using both terrestrial and aquatic

animals as intermediate hosts (in which the parasite completes part of its life cycle) and definitive hosts (in which the parasite reproduces) and the potentially greater egg viability and increased preservation of parasite eggs in wetter soils.

LIMITATIONS AND FUTURE WORK

Differential preservation of parasite eggs in climatically diverse areas of the empire needs to be considered when making comparisons of the parasite diversity in different regions. Relatively little is understood about the specific soil conditions that preserve parasite eggs. Waterlogged conditions have resulted in exceptional preservation of a variety of parasite eggs,¹⁰⁹ likely due to the anaerobic conditions that prevent oxidative decay. The impact of soil acidity on parasite egg preservation has not been studied experimentally, though we may expect that eggs could survive in a wide range of acidities as they need to pass through the gastrointestinal tract, which has a pH ranging from 2 in the stomach to 7 in the small and large intestines. In addition, differential preservation of eggs from different taxa may be contributing to the patterns seen. The general structure of eggshells consists of multiple layers including an outer vitelline layer made up of lipoproteins, a chitinous layer with varying density and orientation of protein fibrils in different parasite species, and an inner lipid layer.¹¹⁰ The chitinous layer that gives the shell strength is expected to be important for preservation, similar to chitin in the exoskeletons of arthropods and fungal spores.¹¹¹ It is generally thought that roundworm and whipworm eggs preserve quite well because they have robust shells. They are found all over the world in various time periods (the oldest recorded human roundworm egg came from Paleolithic France).¹¹² Certain parasites such as hookworm and pinworm are known to have thinner eggshells, resulting in poor preservation.¹¹³ Delicate pinworm eggs have been shown to preserve quite well in coprolites from arid climates, such as that in the western United States, but are not often found in the Old World.¹¹⁴ Other macro- and microorganisms in the soil also contribute to parasite egg preservation.

¹⁰³ Mylona 2018.

¹⁰⁴ Common intermediate hosts for *Diphyllobothrium* sp. in Europe include perch and pike (Scholz et al. 2009, 148).

¹⁰⁵ Curtis 1991, 14; Van Neer et al. 2010.

¹⁰⁶ Kottek et al. 2006.

¹⁰⁷ Izdebski et al. 2016.

¹⁰⁸ Maicher et al. 2019.

¹⁰⁹ Maicher et al. 2017; Ledger et al. 2019; Maicher et al. 2019.

¹¹⁰ Wharton 1980.

¹¹¹ Rinaudo 2006.

¹¹² Bouchet et al. 1996.

¹¹³ Bouchet et al. 2003b.

¹¹⁴ Reinhard et al. 2016, 597; Camacho et al. 2018.

It has been shown that predation by fungi, mites, and other insects can contribute to the loss of parasite eggs over time.¹¹⁵ The presence of these other organisms is complex and can depend on the general ecology of a site and vary in different contexts within a site.

There are other limitations to comparing parasite taxonomic diversity at such a broad scale. First, we must acknowledge that evidence for parasites across the empire comes from a mix of sample types including latrine sediments, sewers, pits, pelvic soil, and mummies that may all preserve eggs differently. Since many more sites have been studied in northern Europe than in the Mediterranean region, this may also skew the results. Additional work from a variety of sites across the empire, and various contexts within these sites, will allow for a more in-depth discussion of parasite diversity regionally. However, with the addition of the material from the two sites in this study, there is now evidence for intestinal parasites from three Roman-period sites in Turkey, one in Greece, and one in Serbia. Only *Giardia duodenalis*, roundworm, and whipworm have been found in these eastern areas of the empire.¹¹⁶ Despite potential differences in preservation, we would expect that parasites that have thick chitinous walls, such as beef/pork tapeworm (*Taenia* spp.), fish tapeworm, and lancet liver fluke, would preserve in similar conditions if they had been present. Further work on how the eggs from different species of parasites preserve will aid in interpretations of past taxonomic diversity globally. Regardless, the results from our studies in the eastern region of the empire suggest that parasite diversity in Roman cities in this region was lower than in provinces north of the Mediterranean. Regional variations in cooking practices, dietary preferences, and climate might have resulted in a lower incidence of zoonotic parasite infections, while population density, sanitation infrastructure, and hygiene practices might have produced conditions that allowed for the presence of fecal-oral pathogens.

CONCLUSIONS

We analyzed samples from two sites in the eastern Roman empire to look for preserved intestinal parasite eggs and cysts. Roundworm eggs were found at both of the sites studied, Sardis and Viminacium; whip-

worm was found at Viminacium. These sites, in combination with recent evidence for *Giardia duodenalis*, roundworm, and whipworm from Kea, Ephesus, and Sagalassos, have revealed the predominance of fecal-oral parasites in Greece, Anatolia, and the Balkans. We have considered explanations for the successful transmission of fecal-oral parasites in the eastern empire, highlighting the role of sanitation infrastructure, latrine sharing and location, and use of human excrement as fertilizer. The presence of these parasites indicates that sanitation provisions and methods for removing human waste were not adequate to stop the spread of fecal-oral parasites in these cities.

As evidence for parasitic infection in the Roman empire grows, it becomes possible to explore variations in parasite species in different regions. As our samples from Turkey and Serbia have shown, fecal-oral parasites were ubiquitous throughout the empire, but the absence of zoonotic parasites demonstrates that infections in Anatolia and the Balkans differed from those in northern Europe. At many sites in the northern provinces, both fecal-oral and zoonotic parasites were present. This indicates that parasitic infections of individuals living in different regions of the empire were varied, and the cultural determinants of these diseases were likely a factor along with local ecology and environment. Possible explanations for these differences include variations in diet and culinary practices, differences in animal management practices, and contrasts in climate between arid regions and well-watered areas. While historical texts inform us of many cultural differences in the empire, paleoparasitological evidence exemplifies the complex interactions that impacted the health and disease of those living within the borders of the empire. Additional studies on both climate and parasite egg preservation are necessary if we wish to better understand regional patterns in parasite diversity across the empire.

Marissa L. Ledger
Department of Archaeology
The Henry Wellcome Building
University of Cambridge
Fitzwilliam Street
Cambridge CB2 1QH
United Kingdom
mll45@cam.ac.uk

Erica Rowan
Department of Classics
Royal Holloway University of London
erica.rowan@rhul.ac.uk

¹¹⁵Morrow et al. 2016.

¹¹⁶Williams et al. 2017; Anastasiou et al. 2018; Ledger et al. 2018.

Frances Gallart Marques
Harvard Art Museums
frances_gallart-marques@harvard.edu

John H. Sigmier
Art and Archaeology of the Mediterranean World
Graduate Group
University of Pennsylvania
jsigmier@sas.upenn.edu

Nataša Šarkić
Department of Biology
Universidad Autónoma de Madrid
nsarkic@gmail.com

Saša Redžić
Institute of Archaeology, Belgrade
sasa.redzic@gmail.com

Nicholas D. Cahill
Department of Art History
University of Wisconsin–Madison
ndcahill@wisc.edu

Piers D. Mitchell
Department of Archaeology
University of Cambridge
pdm39@cam.ac.uk

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