2014

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Published in *International Journal of Paleopathology* 7 (December 2014), pp. 83–87; doi: 10.1016/j.ijpp.2014.08.001

Submitted February 28, 2014; revised August 5, 2014; accepted August 6, 2014; published online August 31, 2014.

Taphonomic considerations of a whipworm infection in a mummy from the Dominican Church of the Holy Spirit, Vilnius, Lithuania

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Abstract

In the present study, the abdominal contents of 10 mummies from beneath the Dominican Church of the Holy Spirit in Vilnius, Lithuania, were examined for the presence of helminth parasites using standard archaeoparasitological techniques. Of the mummies examined, only one individual presented with evidence of parasitism. This individual was infected with both *Trichuris trichiura* and *Ascaris lumbricoides* (5,222 parasite eggs/gram). The conditions of many of the *T. trichiura* eggs suggest that a fortuitously embedded female whipworm decomposed within the individual's gut to release the eggs, as opposed to the eggs actually being passed by the adult helminth. This study highlights a taphonomic issue unique to mummies by demonstrating the differential preservation of parasite eggs existing in various stages of development. Whenever one is not dealing with parasite eggs that have already been passed by the host, as is the case when analyzing intestinal tissues, one must understand that some types of parasite eggs may not be fully formed. It is imperative, as demonstrated by our findings, that researchers have the knowledge to recognize under-developed intestinal helminth eggs in addition to fully formed intestinal helminth eggs from mummy source materials. Together, these findings demonstrate the persistence of these helminth parasites in Europe during the 18th and 19th centuries and represent the first archaeoparasitological evidence from mummies in Vilnius, Lithuania.

Keywords: *Ascaris lumbricoides, Trichuris trichiura,* Lithuania, Mummy, Pathoecology, Vilnius

1. Introduction

Analyses of coprolites, latrines, and mummies from Europe have formed an archaeoparasitological record that reflects the ubiquity of fecal-borne parasitic diseases in this region. This topic is reviewed in other extensive treatises (Bouchet et al., 2003; Leles et al., 2010; Reinhard and Pucu, 2013). Helminths, particularly *Trichuris trichiura* (human whipworm) and *Ascaris lumbricoides* (human mawworm), have been found overwhelmingly in European contexts. Their abundance in the archaeoparasitological record can teach us valuable lessons about the emergence and control of parasitism, if the evidence is properly quantified. Broad-based quantification by Jones (1985) established the importance of quantification in documenting the dispersion of parasites in Medieval contexts. This work was an inspiration for US researchers. Fisher *et al.* (2007) provided a landmark study using quantification on broad scale to document the emergence and control of filth-borne parasitism in Albany, New York.

Quantification methods have been specified for coprolites (Reinhard et al., 1986; Sianto et al., 2005; Fugassa et al., 2011; Jiménez et al., 2012), latrines (Jones, 1985; Reinhard et al., 1986, 2008; Warnock and Reinhard, 1992; Fisher et al., 2007; Florenzano et al., 2012), and skeletons (Fugassa et al., 2006). In contrast to these types of source materials, quantification of parasites from mummies has rarely been done. This is to be expected from studies prior to innovations of quantification methods (Reinhard et al., 1986). However, in recent years researchers have provided such work on mummies (Kumm et al., 2010; Searcey et al., 2013). The importance in quantification is demonstrated by two analyses of Zweeloo Woman, a Dutch bog body. The first study by Paap (1990) revealed the presence of whipworm and

Figure 1. Mummy VD20 with associated abdominal content sample (inset).

ascarids. A second analysis by Searcey *et al.* (2013) showed that the number of eggs in this individual was very low and indicated a symptomless infection. The importance of quantification was also demonstrated by an analysis of the Piraino 1 mummy from Sicily (Kumm et al., 2010). In this instance, quantification revealed evidence of a very heavy infection, which likely provoked symptoms in the individual.

Taphonomy has only recently been recognized as a confounding influence on quantification of eggs per sample and number of individuals infected in a population. Leles *et al.* (2010) were the first to address differential preservation of *Ascaris* eggs in prehistoric American studies. Reinhard and Urban (2003) showed that immature eggs derived from decomposing tapeworms in an intestine have different morphological parameters than mature eggs found in non-mummy contexts. This was also found by Richardson *et al.* (2012) for helminth eggs in dog mummies. Searcey *et al.* (2013) showed that the Zweeloo mummy eggs were less well preserved than the Piraino mummy eggs. These few studies show that archaeoparasitologists should begin documenting the taphonomic factors at work in mummified remains that could impact the presence of eggs and the diagnostic potential of recovered eggs.

It is our goal in this analysis to further explore taphonomic factors on paleoparasitological material in mummy contexts. To that effect, we studied 10 mummies, which are part of a larger study of Lithuanian mummies from a subterranean chamber beneath the Dominican Church of the Holy Spirit of Vilnius (also known as the Lithuanian Mummy Project). This crypt holds mummified and skeletonized human remains of both members of the clergy and laypeople. The site also houses wooden coffins and precious textiles once associated with the bodies. These remains most likely date from the 18th and 19th centuries AD, and belonged to the middle-upper social echelons, according to a widespread European burial custom of locating the dead inside crypts (Frick, 2013). In the fourth decade of the 20th century, students of the local University attempted to sort the remains. However, they did not complete the work, nor were scholars able to locate any written record of their activities, with the exception of two inscriptions on wooden boxes containing collected skeletal elements, as well as some numbers written on the coffins and indicating the different rooms of the crypt.

A report on the mummies was not produced until 1963, when a forensic scientist, Juozas Markulis (1913–1987), briefly analyzed over 500 subjects. Markulis' analyses were aimed at understanding whether the remains were historical or included some 20th century victims. Markulis also looked for evidence of torture, injuries, or other relevant features when conducting these analyses (Markulis, 1963). This report established that most of the corpses were spontaneously mummified, with very few exceptions which bore evidence of evisceration and the use of aromatics. Unaware of his contribution in the field, Markulis initiated mummy studies in Lithuania.

It should be said that throughout history, the mummies were manipulated. Manipulations of these individuals can be attributed to the site being used by Napoleonic soldiers in 1812, the continual deposition of dead bodies, and even as a bomb shelter during the Nazi period. Most of the remains had been placed in one single cellar, and a glass window was installed in the 1960s, which triggered a change in air flow. This change in air flow caused most of those remains to begin to decompose. Only about 23 mummies could be retrieved as part of the Lithuanian Mummy Project.

At the request of the Church officials, an investigation enabling the documentation, study, and curation of this historic material commenced in 2011 with the study of such mummies consisting of 15 adults, 8 sub-adults and a number of isolated body parts (Jankauskas and Piombino-Mascali, 2012; Panzer et al., 2013; Piombino-Mascali et al., 2014). These are the only surviving mummies. Following the external inspection, each body was carefully recorded and tissue and bone samples were obtained from those subjects who displayed loss of substance, either determined by natural decay or other post-depositional changes. Tissue samples relevant to archaeoparasitological examinations were collected from 10 of the individuals from this site and submitted to the palynology/pathoecology laboratory of the School of Natural Resources at the University of Nebraska for analysis.

2. Materials and Methods

Samples were studied at the palynology/pathoecology laboratory of the School of Natural Resources at the University of Nebraska for analysis in 2014. This is a filtered air, positive pressure facility that is free of contaminants smaller than a micrometer. The samples were sealed in sterile ziplock bags. The closures for all bags were intact upon arrival. To prevent DNA contamination of material not used in this analysis, gloves were worn by the lab personnel who handled the specimens.

The focus of this study is a sample of abdominal contents from an 18th to 19th century subadult of undetermined sex (VD20). Though the abdominal contents from 10 mummies were analyzed, only this individual presented with evidence of parasitism. The methods employed for individual VD20 were consistent with the methods used to analyze the other nine individuals that presented with no evidence of parasitism. VD20 itself was only half of a body, comprised of an area extending from the abdomen to the feet (Figure 1). A laboratory analysis number (Lab #11) was assigned to the sample and it was photographed using a SONY Cybershot 18.5 megapixel camera

Table 1. VD20 (Lab #11) microfossil counts and values used for quantification.

Microfossil type	Microfossils counted	*Lycopodium* counted	*Lycopodium* added	Subsample weight (gram)	Microfossil concentration
A. lumbricoides	5	92	25,000	3.07	442 eggs/gram
T. trichiura	54	92	25,000	3.07	4,779 eggs/gram
Total parasite eggs	59	92	25,000	3.07	5,222 eggs/gram

before it was opened. A subsample was taken from the sample and weighed (Figure 2 and Table 1). The subsample was then placed into a 50 mL plastic centrifuge tube and covered in a 0.5% trisodium phosphate solution. The subsample was left to rehydrate in this solution for 48 h.

Parasite egg concentration, quantified using the *Lycopodium* method, was originally developed by Warnock and Reinhard (1992). It has subsequently become standard among archaeo- and paleoparasitologists (Martinson et al., 2003; Reinhard and Urban, 2003; Santoro et al., 2003; Sianto et al., 2005; Reinhard et al., 2008, 2012; Kumm et al., 2010; Fugassa et al., 2011; Jiménez et al., 2012; Searcey et al., 2013). Following rehydration, the intestinal contents were moved to 300 mL glass beakers and treated with two *Lycopodium* spore tablets (Batch # 124961), each containing approximately 12,500 spores. Tablets were dissolved in hydrochloric acid (HCl) in sterilized 50 mL plastic beakers prior to being added to the subsample (Stockmarr, 1971). The beakers were rinsed three times to ensure that all spores were transferred to the abdominal sample. The subsample was subsequently disaggregated using a magnetic stirrer and clean stir bar.

The subsample was then rinsed with a jet of distilled water over a 250 micron mesh screen into a 600 mL glass beaker. Following screening, macroscopic remains were transferred from the mesh screens onto sterile filter paper, labeled, and left to dry. The dried macroremains were later examined using an Olympus SZ-PT dissection microscope.

The liquid reserved from screening was concentrated via repeated centrifugation. The concentrated remains were analyzed for the presence of parasites and starch. To perform these microscopic analyses, examination slides were created by mixing a small amount of concentrated subsample material with a drop of glycerin onto a clean glass microscope slide with a 22 mm × 22 mm cover slip. Slides were examined via light microscopy using a Nikon compound microscope equipped with Infinity Image Capture hardware. Photographs were taken using the associated software.

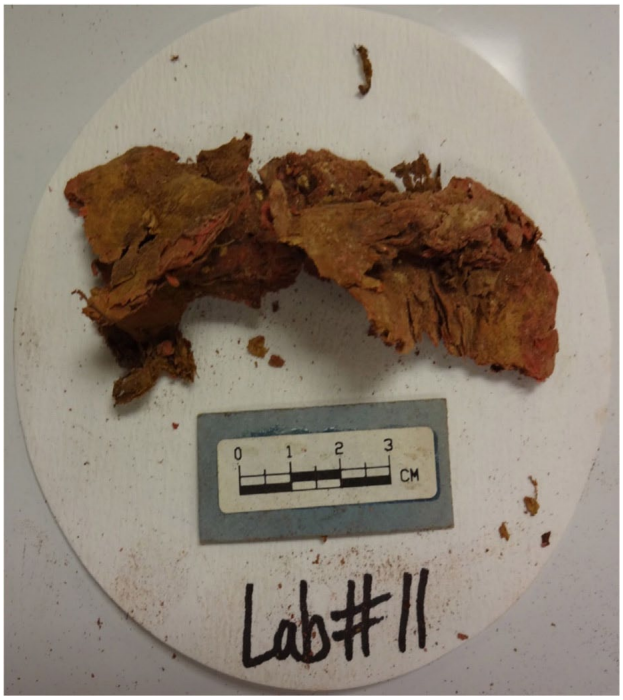

Figure 2. Subsample of VD20 abdominal contents.

Parasite egg concentration values were calculated using the following formula: egg concentration = $[(p/m) \times a]/v$, where p was the number of parasite eggs counted, m was the number of *Lycopodium* spores counted, a was the number of *Lycopodium* spores added to the sample, and v was the total weight of the sample prior to rehydration.

After parasite and starch analyses, half of the subsample material was archived and the other half was processed for palynological analysis. The sample was archived using standard archaeoparasitological archival techniques and was deposited in the collection of mummy parasite preparations housed in the palynology/pathoecology laboratory, School of Natural Resources, University of Nebraska–Lincoln. The starch, pollen, and macrofossil analyses from this sample are on-going.

3. Results

After examining the 10 samples, only one individual was found to be host to parasitic infections. The other nine samples tested negative for helminthiasis. The results from the parasitized individual (VD20 [Lab #11]) are presented below.

Initial observations of Lab #11 revealed that the sample consisted of intestinal material as well as red paint. The origin of the paint is unclear, but, as much as possible, we avoided including paint when taking the subsample. The original sample was friable and fragmented so that it was easy to obtain a subsample. The subsample weighed 3.07 g and appeared as a dark, brown, opaque mass after rehydration. It turned the rehydration solution opaque and dark brown. This coloration following rehydration is indicative of remains that are highly organic in nature. This is to be expected from abdominal materials. Samples that are comprised largely of inorganic material appear yellow and translucent following rehydration.

Microscopic analysis revealed evidence of both trichuriasis and ascariasis in the individual (Table 1). Parasite egg concentrations were higher for *T. trichiura* (4,779 eggs/gram) than for *A. lumbricoides* (442 eggs/gram). The fact that these samples came from an intestinal sample, along with substantial egg concentrations, shows that there was a true infection in the individual examined (VD20).

The *A. lumbricoides* eggs seemed to be well preserved in most cases although some of the eggs encountered were folded. None of the eggs demonstrated evidence of fissures or decortication of the mammillated coat. The lack of these alterations indicates that the abdominal environment was well suited for preservation of the ascarid eggs and that our methods were not damaging to the microfossils. On the other hand, the eggs of *T. trichiura* were not as well preserved as were the ascarid eggs (Figure 3). Many of the whipworm eggs were without their polar plugs or any embryonic contents.

Additionally, many of the whipworm eggs were recovered in couplets, triplets, and a conglomeration of nine eggs (Figures 4 & 5). The significance of these recoveries will be discussed at length in the following section.

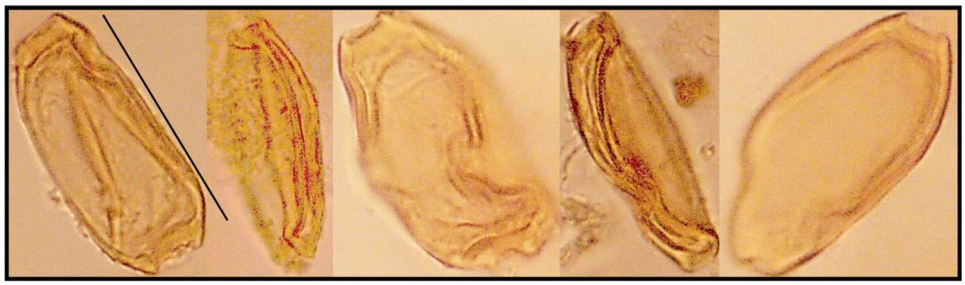

Figure 3. Crushed, folded, and otherwise damaged eggs of *T. trichiura* recovered from VD20 (scale bar = 50 µm).

Figure 4. Cluster of three *T. trichiura* eggs recovered from VD20 (scale bar = 50 µm).

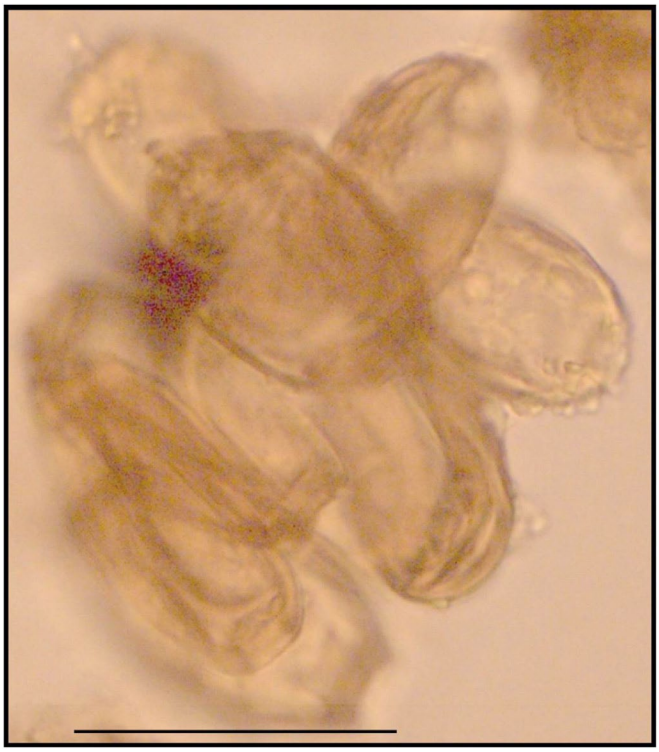

Figure 5. A cluster of nine *T. trichiura* eggs recovered from VD20 (scale bar = 50 µm).

4. Discussion

The results of archaeoparasitological analysis show that VD20 represents a case of helminthiasis in a historic Lithuanian mummy. This individual was infected with two different species of helminths (*A. lumbricoides* and *T. trichiura*) as indicated by parasite egg concentrations (442 eggs/g and 4,779 eggs/gram, respectively). To our knowledge, this is the first case of either ascariasis or of trichuriasis reported from a Lithuanian mummy. The fact that only one of ten individuals was infected indicates that infection may not have been common in historic Lithuania. As discussed by Reinhard and Pucu (2013) helminth infections were a universal problem from the Neolithic through the Renaissance in other parts of Europe. These results suggest that there may have been sanitation issues among the Vilnius community between the 18th and 19th centuries or that the climate may have been conducive to the life cycle of these helminths.

This research also brings up the interesting taphonomic point of differential parasite egg preservation from abdominal contexts. The preservation of eggs in mummies ranges from pristine (Kumm et al., 2010) to excellent (Seo et al., 2007; Shin et al., 2009), deformed (Reinhard and Urban, 2003) to degraded (Searcey et al., 2013). Some whipworm eggs documented by Kumm et al. (2010) were fully embryonated which is unexpected in mummy contexts.

Poor preservation of eggs in mummies makes a great deal of sense from the perspective of intestinal contents, and especially intestinal sections. These remains may contain decomposed adult helminths with incompletely formed eggs in utero. The community of mummy researchers has been slow to recognize this variation. We believe that it is necessary to acknowledge this and plan for repeat analysis of negative samples.

The *T. trichiura* eggs were thin-walled and variously damaged or deformed in comparison to eggs of the same species recovered from mummies from other sites (Aspöck et al., 1996; Martinson et al., 2003; Seo et al., 2007; Shin et al., 2009; Kumm et al., 2010). The polar plugs may not have been missing, but rather may not have yet been formed. In other words, these appear to be immature eggs of the sort found within a whipworm uterus.

Additionally, the recovery of multiple eggs in couplets, triplets, and a conglomeration of nine eggs suggest that some of these eggs were from a gravid female *T. trichiura*, which burst open sometime during our analyses releasing eggs that were

not fully developed (Figures 4 & 5). We are certain that the clustering of eggs is not due to the fluid dynamics of organic matter in the samples. The remains were thoroughly rinsed and other microfossils, such as pollen, were not clustered. The clustering, therefore, is a specific phenomenon relating to the whipworm eggs. Also, in numerous analyses carried out in our labs, we have never before encountered clustering such as this (Martinson et al., 2003; Santoro et al., 2003; Fugassa et al., 2006, 2011; Reinhard et al., 2008; Kumm et al., 2010; Jiménez et al., 2012; Rácz et al., 2014).

The observed clustering is analogous to the situation reported by Reinhard and Urban (2003) regarding the taphonomy of fish tapeworm eggs in Chinchorro mummy context and Richardson *et al.* (2008) for Peruvian mummified dogs. They asserted that the fact that immature eggs, as well as mature eggs, were trapped within the mummy resulted in a taphonomy unique to mummies in which the pliable, incompletely formed eggs can be recovered. The apparent poor preservation of immature eggs is due to the fact that they were never completely formed. We believe that in VD20, we fortuitously rehydrated material that included fragments of a decomposed female whipworm containing eggs in various forms of maturation.

As noted by several authors, these parasites infected people throughout prehistory in the region (Leles et al., 2010; Reinhard and Pucu, 2013). In the general region, two famous bog bodies were discovered before World War II that proved positive for *A. lumbricoides* and *T. trichiura* (Szidat, 1944). These were found in the western part of East Prussia which is currently part of Poland. The bog bodies are a 12–14 year-old Iron Age female from Dröbnitz and an older adult male from Karwinden. Analyses of these bodies and other contributions by Szidat have been summarized by Dönges (1994). These discoveries show a continuity in parasitism in eastern Europe from the Iron Age onward. The data from our study further support that infections with whipworm and mawworm were present in this part of the world for an extended period of time. This study also highlights unique taphonomic issues that arise with the archaeoparasitological analysis of mummies.

Acknowledgments — The CAPES Ciência Sem Fronteiras sponsored the collaborators from FIOCRUZ. The authors would like to thank the University of Nebraska Undergraduate Creative Activities and Research Experiences Program (UCARE) for lab support. The scientific study of these mummies was kindly supported by a Lithuanian State scholarship awarded to one of the authors (Dario Piombino-Mascali).

References

Aspöck, H., Auer, H., Picher, O., 1996. *Trichuris trichiura* eggs in the Neolithic glacier mummy from the Alps. *Parasitol. Today* 12, 255.

Bouchet, F., Guidon, N., Dittmar, K., Harter, S., Ferreira, L.F., Chaves, S.M., Reinhard, K.J., 2003. Parasite remains in archaeological sites. *Mem. Inst. Oswaldo Cruz* 98,47–52.

Dönges, J., 1994. Centenary biographical note: Lothar Szidat 1892–1973. *Int. J. Parasitol.* 24, 163–164.

Fisher, C.L., Reinhard, K.J., Kirk, M., DiVirgilio, J., 2007. Privies and parasites: The archaeology of health conditions in Albany, New York. *Hist. Archaeol.* 41 (4), 172–197.

Florenzano, A., Mercuri, A.M., Pederzoli, A., Torri, P., Bosi, G., Olmi, L., Rinaldi, R.,Mazzanti, M.B., 2012. The significance of intestinal parasite remains in pollen samples from Medieval pits in the Piazza Garibaldi of Parma, Emilia Romagna, Northern Italy. *Geoarchaeology* 27 (1), 34–47.

Frick, D., 2013. *Kith, Kin, & Neighbors: Communities & Confessions in Seventeenth-Century Wilno.* Cornell University Press, Ithaca, New York.

Fugassa, M.H., Araújo, A., Guichon, R.A., 2006. Quantitative paleoparasitology applied to archaeological sediments. *Mem. Inst. Oswaldo Cruz* 101, 29–33.

Fugassa, M.H., Reinhard, K.J., Johnson, K.L., Vieira, M., Araújo, A., 2011. Parasitism of prehistoric humans and companion animals from Antelope Cave, Mojave County, Northwest Arizona. *J. Parasitol.* 97 (5), 862–867.

Jankauskas, R., Piombino-Mascali, D., 2012. The Lithuanian Mummy Project: bioanthropological and paleopathological investigation of the human remains found in the Holy Spirit Dominican Church, Vilnius. *Paleopathol. Newsl.* 159, 12.

Jiménez, F.A., Gardner, S.L., Araújo, A., Fugassa, M.H., Brooks, R.H., Rácz, E., Reinhard,K.J., 2012. Zoonotic and human parasites of inhabitants of Cueva de Los Muertos Chiquitos, Rio Zape Valley, Durango, Mexico. *J. Parasitol.* 98 (2), 304–309.

Jones, A.K., 1985. Trichurid ova in archaeological deposits: Their value as indicators of ancient faeces. In: *Palaeobiological Investigations. Research Design, Methods and Data Analysis*, Symposia of the Association for Environmental Archaeology B 5., pp. 105–115.

Kumm, K., Reinhard, K.J., Piombino-Mascali, D., Araújo, A., 2010. Archaeoparasitological investigation of a mummy from Sicily (18th–19th century AD). *Anthropologie* 48, 177–184.

Leles, D., Reinhard, K.J., Fugassa, M., Ferreira, L.F., Iniguez, A.M., Araújo, A., 2010. A parasitological paradox: Why is ascarid infection so rare in the prehistoric Americas? *J. Archaeol. Sci.* 37, 1510–1520.

Markulis, J., 1963. Vilniaus Domininkonų bažnyčios požemiuose rastųjų palaikųteismo medicininio tyrimo aktas. Valstybinis V. Kapsuko universitetas, Vilniuje.

Martinson, E., Reinhard, K.J., Buikstra, J.E., Dittmar, K., 2003. Pathoecology of Chiribaya parasitism. *Mem. Inst. Oswaldo Cruz* 98, 195–205.

Paap, N.A., 1990. Parasieten. In: van der Sanden, W.A.B (ed.), *Mens en moeras–veenlijken in Nederland van de bronstijd tot en met de Romeinsetijd.* Drents Museum, Assen, the Netherlands, pp. 162–167.

Panzer, S., Tamošiūnas, A., Valančius, R., Jankauskas, R., Piombino-Mascali, D., 2013. Radiological evidence of rickets in a Lithuanian child mummy. *Rofo* 185,670–672.

Piombino-Mascali, D., Kozakaitė, J., Tamošiūnas, A., Valančius, R., Panzer, S., Jankauskas, R., 2014. Skeletal pathological conditions of Lithuanian mummies. *Papers Anthropol.* 23 (1), 118–126.

Rácz, S.E., Pucu, E., Jensen, E., Mostek, C., Morrow, J.J., Van Hove, M.L., Bianucci, R.,Willems, D., Heller, F., Reinhard, K.J., 2014. Parasitology in an archaeological context: Analysis of medieval burials in Nivelles, Belgium. *J. Archaeol. Sci.* (in press).

Reinhard, K.J., Pucu, E., 2013. Comparative parasitological perspectives on paleoepidemiological transitions: Americas and Europe. In: Zuckerman, M.K. (ed.), *Moving the Middle to the Foreground: Interdisciplinary Approaches to Examining the Second Epidemiological Transition.* Wiley-Blackwell Publishing, Hoboken, NJ, pp. 311–326.

Reinhard, K., Urban, O., 2003. Diagnosing ancient diphyllobothriasis from Chinchorro mummies. *Mem. Inst. Oswaldo Cruz* 98, 191–193.

Reinhard, K.J., Araújo, A., Sianto, L., Costello, J.G., Swope, K., 2008. Chinese liver flukes in latrine sediments from Wong Nim's property, San Bernardino, California: Archaeoparasitology of the Caltrans District Headquarters. *J. Parasitol.* 94,300–303.

Reinhard, K.J., Johnson, K.L., LeRoy-Toren, S., Wieseman, K., Teixeira-Santos, I., Vieira,M., 2012. Understanding the pathoecological relationship between ancient diet and modern diabetes through coprolite analysis: A case example from Antelope Cave, Mojave County, Arizona. *Curr. Anthropol.* 53, 506–512.

Reinhard, K.J., Confalonieri, U.E., Hermann, B., Ferreira, L.F., Araujo, A.J., 1986.Recovery of parasite remains from coprolites and latrines: Aspects of paleoparasitological technique. *Homo* 37, 217–239.

Richardson, D.J., Guille, S., Beckett, N.R., Kyle, W., Conlogue, G., Harper-Beckett,K., 2012. Archaeohelminthology of the Chiribaya Shepherd, *Canis familiaris* (700–1476 AD) from southern Peru. *Comp. Parasitol.* 79, 133–137.

Santoro, C., Vinton, S.D., Reinhard, K., 2003. Inca expansion and parasitism in the Lluta Valley: Preliminary data. *Mem. Inst. Oswaldo Cruz* 98, 161–163.

Searcey, N., Reinhard, K.J., Gardner, S.L., Egarter-Vigl, E., Maixner, F., Piombino-Mascali, D., Zink, A., Van Der Sanden, W., Bianucci, R., 2013. Parasitism of the Zweeloo Woman: Dicrocoeliasis evidenced in a Roman Period bog mummy. *Int. J. Paleopathol.* 3, 224–228.

Seo, M., Guk, S.M., Kim, J., Chai, J.Y., Bok, G.D., Park, S.S., Oh, C.S., Kim, M.J., Yil, Y.S.,Shin, M.H., Kang, I.U., Shin, D.H., 2007. Paleoparasitological report on the stool from a Medieval child mummy in Yangju, Korea. *J. Parasitol.* 93, 589–592.

Shin, D.H., Lim, D.S., Choi, K.J., Oh, C.S., Kim, M.J., Lee, I.S., Kim, S.B., Shin, J.E., Bok, G.D.,Chai, J.Y., Seo, M., 2009. Scanning electron microscope study of ancient parasiteeggs recovered from Korean mummies of the Joseon Dynasty. *J. Parasitol.* 95,137–145.

Sianto, L., Reinhard, K.J., Gonc¸alves, M.L.C., Araújo, A., 2005. The finding of Echinos-toma (Trematoda: Digenea) and hookworm eggs in coprolites collected from aBrazilian mummified body dated 600–1200 years before present. *J. Parasitol.* 91,972–975.

Stockmarr, J., 1971. Tablets with spores used in absolute pollen analysis. *Pollen et Spores* 13, 615–621.

Szidat, L., 1944. Über die Erhaltungsfähigkeit von Helmintheneiern in Vor- und Frühgeschichtlichen Moorleichen. *Z. Parasitenkd.* 13, 265–274.

Warnock, P.J., Reinhard, K.J., 1992. Methods for extracting pollen and parasite eggs from latrine soils. *J. Archaeol. Sci.* 19, 261–264.