



## *Toxocara* eggs in an 18th century Franciscan from Portugal. The challenge of differentiating between parasitism and chance in Paleoparasitology



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### ABSTRACT

In 2005, an adult male was excavated in the cloister of the former Convent of the Holy Spirit, in the Franciscan Province of Holy Mary of Arrábida, Lisbon district. From the anterior part of the sacrum, a darker organic agglomeration was collected and studied for intestinal parasites. Samples were rehydrated with *Lycopodium* tablets in a Na<sub>3</sub>PO<sub>4</sub> 5% solution for 72 h, followed by the swirl technique. Organic material was concentrated at 2500 rpm. At least 20 slides of each sample were examined using a light/polarized microscope. A control sample from outside the pelvis revealed no biological remains. A sample collected inside the pelvic girdle was positive for pollen grains, other plant remains and *Toxocara* eggs, perhaps *T. cati* (2766 eggs/gram sediment). This finding, although exciting, cannot be explained in a simple way because humans are not definitive hosts for *Toxocara* species. Ingestion of feces-contaminated food or water, geophagy, or true infection are hypotheses considered in this study, which demonstrates the difficulty of interpreting the presence of animal parasites in human remains. This is the first time *Toxocara* eggs are found associated with human remains.

### 1. Introduction

Since the beginning of paleoparasitology, animal parasites have been found associated with human remains, which usually indicates zoonoses and false parasitism (Sianto et al., 2009). These situations are differentiated mostly on the basis of the parasite's specificity and biological cycle. In most cases, however, this differentiation is hampered by the absence of symptomatology and other clinical data on individuals. In these situations, a combination of laboratory diagnosis, historical resources, and knowledge of biological and cultural habits may help in the interpretation of parasite-host relationships. Sometimes the presence of a parasite may evidence domestication of plants and animals (Reinhard et al., 2013), unusually food habits (Reinhard, 2006; Sianto et al., 2012); or local food shortages (Le Bailly et al., 2007; Reinhard, 2008). In this article we present possible explanations for *Toxocara* eggs found in pelvic sediments of an 18th century Franciscan friar from the Convent of the Holy Spirit in Mealhada, Lisbon district.

Franciscanism was established in Portugal in 1217. Friars Zacarias

and Gualter were sent to Portugal by Saint Francis of Assisi, charged with the task of developing religious propaganda aimed at the construction of new convents (Moreno, 1995). The first Portuguese convents were built in Leiria in 1232 and Porto in 1233 (Moreira, 1996). The first Franciscan presence in Loures dates from the 13th century, where the convent of the Divine Holy Spirit held the Ordem Terceira de S. Francisco. This convent was destroyed by the earthquake of 1531. At the site of its ruins, a small hermitage, with a small hostel and hospital, was built in 1541. In 1838, it was completely destroyed by the flood of the Trancão river, a tributary of the Tagus river (Mendes Leal, 1909). The convent of the Holy Spirit was built in Mealhada, close to Loures, in 1574 to welcome the Franciscan Friars of the Province of Santa Maria da Arrábida, who were the most austere and humble of the remaining Franciscan provinces in terms of rule and discipline. The convent was responsible for integrating and training novices and for teaching Latin grammar, reading and writing; it housed just over 10 friars and novices. Additionally, the hostel was integrated into the region's health itinerary, receiving patients on their way to the Hospital

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of All Saints in Lisbon (Gomes, 1982).

The monks possessed humble garments – wool habits (burel) – and only the elderly and infirm could wear shoes, which some refused as a form of penitence. Produced handmade pieces were not for sale, they were only for divine worship in the chapels. Religious duties consumed much of the monks' time and included prayers for the deceased, funerals, processions, mental prayer and confessions of two-and-a-half-hours daily. The rest of their time was filled with domestic activities such as washing the habits and other clothes of the religious community, cleaning the convent, transporting water to the service areas, providing assistance to the hostel and preparing meals (Estatutos da Provincia de Santa Maria da Arrabida, 1698).

## 2. Methodology

The archaeological intervention in the Convent of the Holy Spirit occurred in 2005. The cloister is located southwest of the chapel and presents a single floor quadrangular plan with an open gallery containing a section of pillars and a quadrangular column in each corner. The original plan suffered alterations; however, the details are not known. It is coated with tiles depicting secular themes, dating from the 2nd half of the 18th century. There is cover on the vault. Throughout the covered gallery are sixteen graves, arranged lengthwise and covered by two individual calcareous lids. Oval pits dug into loamy dark brown soil were used to receive the burials. Each grave received more than one individual, with only the most recent being preserved because the lack of space forced the reuse of graves, typical at the time (Silva and Antunes-Ferreira, 2005).

Burial 3 (Fig. 1) was the only one evaluated for the presence of parasites. It is a primary inhumation identified under slab 16 in the northwest of the cloister. It is a simple grave that was reused, as indicated by the presence of bones in the sediment. The skeleton was

moderately preserved, with fragile and brittle bones, and the spongy bones were almost reduced to powder (Silva and Antunes-Ferreira, 2005). The individual was in the supine position with the hands on the abdomen and legs extended. The head did not droop. The assumed direction was southwest-northeast, with the head southwest. Some nails were found, indicating deposition in a wooden coffin (Silva and Antunes-Ferreira, 2005). There were no associated assets. However, considering the collected artifacts in the remaining graves of the cloister, we can situate this burial between the 2nd half of the 17th century and the 18th century.

During the delimitation of the skeleton, a darker organic agglomeration was identified in the anterior part of the sacrum, which, based on appearance and localization, was identified as fecal matter from the individual. The greenish pasty material did not present any smell, and it was collected in a sterilized plastic bag. Another sample from the surrounding soil of the skeleton was also collected as a control. Paleoparasitological study was conducted using sediment samples recovered both from the pelvis and surrounding area of the skeleton. Three grams from a total of 21 g taken from the pelvic area, and three grams of the surrounding soil, were rehydrated in 0.5% trisodium phosphate solution (Callen and Cameron, 1960) with commercial *Lycopodium* spore tablets batch 12542. After 3 days, the samples were homogenized with a glass rod and then the swirl technique (Reinhard et al., 2008) was applied to remove sand from the samples. For analysis, at least 20 slides of each sample were inspected at 100–400 x magnifications using a Nikon E200 light/polarized microscope with a Lumenera® camera. Pictures and measurements were taken with Image Pro® Express Software. For the quantification of the eggs and dietary remains per gram of sediment, the formula adapted from Maher (Maher, 1981) was used:  $Mpg$  (microfossils/gram of sediment) = ((microfossils counted/spores counted) x spores added)/sediment weight). The identification of dietary content was conducted only in microscopic remains.

Sex and age of the individual were determined considering the morphological characteristics of the pelvis and skull (Bruzek, 2002; Buikstra and Ubelaker, 1994; Walker, 2008). Paleopathological analysis was performed by macroscopic observation.

## 3. Results and discussion

The age at death assessment suggests that the individual, a male, was older than 30 years of age because of the closure of all epiphyses, including the clavicle sternal end (Buikstra and Ubelaker, 1994; Cunha et al., 2009). The outer sternal end of the 4th rib, auricular surface and pubic symphysis were destroyed and therefore it was impossible to refine the age to death of the individual. The height calculated from the length of the left fibula (339 mm) is 162.14 cm  $\pm$  6.76 cm (Olivier et al., 1978). Paleopathological analysis revealed the loss of at least 15 teeth ante mortem (12 in the mandible and 3 in the upper jaw). Remaining teeth (n = 9) were located only in the upper jaw and presented moderate occlusal wear and severe tartar (especially in the posterior dentition). There is no indication of cribra orbitalia or cribra crania, but linear hypoplasia of tooth enamel was identified in both incisors and canines present. Linear hypoplasia may reveal stress episodes during dental crown formation, being associated with systemic imbalances such as malnutrition, infections, parasitosis, physical and psychological trauma that occur during the growth period of the non-adult individual (Goodman and Martin, 2002; Goodman and Rose, 1991; Larsen, 2002; Powell, 1988; Skinner and Goodman, 1992). In the appendicular skeleton, osteoarthritis was identified, and spinal osteoarthritis is present in the cervical vertebrae. Indirect age indicators such as tooth loss and osteoarthritis in some joints suggest that the individual could be elderly. No bone changes consistent with infectious conditions and injuries were observed.

The sediment sample taken for control did not reveal any parasitological or dietary remains. The sediment collected from the pelvis



Fig. 1. Burial 3 exhumed from the cloister.

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