



# First study of fossil rodent middens as source of paleoparasitological evidences (northwestern Patagonia, Argentina)

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## ABSTRACT

The present paper reports the first paleoparasitological results obtained from coprolites of fossil rodent middens and demonstrates the potential of rodent middens as a source of paleoparasitological evidences in South America. Ten fossil rodent middens from northwestern Patagonia, Argentina, were studied. Five coprolites of each midden were fully processed, rehydrated, homogenized, subjected to spontaneous sedimentation, and examined through light microscopy. Eight of the 10 examined rodent middens contained parasite eggs. The eggs of parasites were assigned to *Heteroxynema* (*Cavioxyura*) *viscaciae* Sutton & Hugot, 1989 and *Helminthoxys* sp. (Nematoda: Oxyuridae), *Trichuris* sp. (Nematoda: Trichuridae) and one unidentified nematode. Fossil rodent middens were assigned to *Lagidium viscacia* (Caviomorph: Chinchillidae). The excellent preservation of parasite remains in coprolites from fossil rodent middens provided an opportunity to perform paleoparasitological inferences. The results of this paper demonstrate that fossil rodent middens offer an excellent opportunity for the recovery of parasite remains for future paleoparasitological studies in the southwest of South America.

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## 1. Introduction

Rodent middens are accumulations of organic debris (plant macrofossils, coprolites, pollen) and sediment, encased in hardened urine preserved in rockshelters, caves and crevices [1]. When water from rodent urine is evaporated, these accumulations hardened, allowing the preservation of the deposited materials which can remain intact for several thousand years [2].

Studies of South American fossil rodent middens have been mainly focused on the analysis of pollen and plant macrofossil records. Such studies provided fundamental paleoecological and paleoclimatological evidence from arid and semi-arid regions of South America for the late Quaternary (last 50 ka) [2–8]. Midden agents in South America includes rodent genera such as *Lagidium* spp. (Chinchillidae), *Phyllotis* spp. (Cricetidae), *Abrocoma* spp. (Abrocomidae), *Microcavia* spp. (Caviidae), *Octomys* sp. and *Octodontomys* sp. (Octodontidae) [2]. Rodent middens have become a valuable archive in arid and semi-arid regions of South America due to their multiproxy evidence which allows to address questions in diverse areas of knowledge such as botany, zoology,

ecology, biogeography, archeology, climatology, molecular biology, among others.

Paleoparasitological studies have been traditionally applied to coprolites, mummified remains and latrines which are the most common sources of evidences [9]. During the past few years, several samples of rodent coprolites collected from archeological and paleontological sites from Patagonia were examined for parasites [10–16] providing important insights into the biodiversity of parasites in ancient times. In that sense, the presence of rodent parasites in fossil middens would provide useful and valuable information, as rodent paleoparasitological data can be used from several point of views [17]. Despite their unique conditions for preservation in arid and semi-arid settings from South America, fossil rodent middens have not been used for paleoparasitological purposes.

The present paper reports, therefore, the first paleoparasitological results obtained from coprolites of fossil rodent middens from northwestern Patagonia, Argentina demonstrate the potential of rodent middens as a source of paleoparasitological evidences in South America.

## 2. Material and methods

Northwestern Patagonia is located on the lee side of the Andes and the adjacent volcanic fields and plateaus between 35°–39°S. The region

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is characterized by a west–east climatic gradient from humid conditions along the Andes and dry conditions at the easternmost tip. Vegetation distribution follows the west–east climatic and environmental gradient ranging from the forest and the grass steppes at the lee side of the Andes to the shrub steppes developing on volcanic fields and plateaus [18,19].

The Huenul locality, where rodent middens were collected, is located near the grass steppe– shrub steppe (locally known as Monte) ecotone, at the easternmost tip of the climatic gradient. It is close to Cueva Huenul 1 archeological site (CH1) (36°56′45″S, 69°47′32″W) which presents excellent preservation conditions of archeological and paleontological deposits [20,21].

Ten fossil rodent middens were collected at Huenul site and specifically together to CH1 and within a 200 m-far gorge from CH1 (Fig. 1; Table 1). Eight of the ten middens were 14C dated at LATYR (Laboratorio de Tritio y Radiocarbono, Argentina) and calibrated with the Southern Hemisphere curve (SHCal13; Hogg et al., 2013) from CALIB 7.0.2 program (Table 1). Five coprolites from each rodent midden were examined for parasites. Coprolites were inventoried and processed individually. The examination of the external shape of feces was conducted according to Chame [22] and Jouy-Avantin [23]. Each coprolite was fully processed by rehydration in a 0.5% water solution of tris-sodium phosphate (TSP) in a glass tube for a week, followed by homogenization, processed by spontaneous sedimentation [24] and preserved in 70% ethanol. Ten slides were prepared from each coprolite, along with the addition of one drop of glycerin to each slide, and were examined using light microscopy. Eggs of parasites were measured and photographed at 400× magnification. Broken eggs were discarded.

### 3. Results

Coprolites were dark brown and cylindrical, with smooth surfaces (Fig. 2). Average measurements of feces (N = 50) were 10.3 ± 1.1 mm long by 3.7 ± 0.7 mm wide.

Eight of the ten rodent middens examined contained parasite eggs (Table 1). Sixteen coprolites contained eggs of nematodes. Eggs of parasite were assigned to *Heteroxynema* (*Cavioxyura*) *viscaciae* Sutton & Hugot, 1989 and *Helminthoxys* sp. (Nematoda: Oxyuridae), *Trichuris* sp. (Nematoda: Trichuridae) and one unidentified nematode.

Eggs of *H. viscaciae* (Fig. 3), with single thick wall and with a rounded pole and the other sharp, without operculum, were found in 5 coprolites from 4 middens. Average egg measurements (N = 5) were 137.5 to 152.5 (143.5 ± 5.47) µm long by 62.5 to 67.5 (65.63 ± 2.39) µm wide.

Asymmetrical and yellowish eggs, with only one side convex and thin and smooth wall, with a dark-yellow embryonic mass, were assigned to *Helminthoxys* sp. (Fig. 4) and were found in 6 coprolites from five middens. Average egg measurements (N = 20) were as follows: 100.0 to 112.5 (106.9 ± 4.12) µm long by 42.5 to 55.0 (48.63 ± 3.19) µm wide.

Eggs of *Trichuris* sp. (Fig. 5) were observed in 4 coprolites from 4 different rodent middens. These eggs were lemon shaped, with a smooth surface and polar plugs. Measurements of eggs were different between middens, and were attributed to two different *Trichuris* species. Average egg measurements (N = 6) from one of the middens were as follows: 70.0 to 77.5 (74.17 ± 2.58) µm without plugs and 77.5 to 80.0 (79.00 ± 1.37) µm with plugs long by 37.5 to 40.0 (38.5 ± 1.37) µm wide. From 2 middens only one egg in each one were found. Measurements were 55 µm without plugs and 65 µm with plugs long by 42.5 µm wide and 60 µm without plugs long by 32.5 µm wide. Two *Trichuris* eggs were found in another midden, their measurements were 52.5 and 57.5 µm without plugs and 60 and 62.5 µm with plugs by 30 µm wide.

Two oval and embryonated egg belong to an unidentified nematode were found in one coprolite, the measurements were 45 µm by 40 µm (Fig. 6).

### 4. Discussion

Species of oxyurid nematodes are monoxenic parasites that live in the posterior third of the digestive tract of various vertebrates and arthropods [25]. Oxyuroidea from vertebrates can be grouped into 3 families: Pharyngodonidae, Oxyuridae and Heteroxynematidae [26]. Heteroxynematidae includes nematodes that evolved in sciromorph, caviomorph and miomorph mammals. *Heteroxynema viscaciae* (Heteroxynematidae) is a parasite found in the caecum and large intestine from *Lagidium viscacia* from Chubut Province, Argentina, first described by Hugot and Sutton [27]. It was also observed by Foster



Fig. 1. Photographs showing a. Cueva Huenul 1 (CH1); b. gorge close to CH1; c. and d. rodent middens collected at Huenul site.

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