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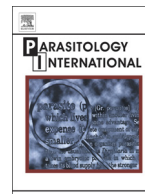
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Parasitology International

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# New record of anoplocephalid eggs (Cestoda: Anoplocephalidae) collected from rodent coprolites from archaeological and paleontological sites of Patagonia, Argentina



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## ARTICLE INFO

### Article history:

Received 29 October 2012

Received in revised form 17 January 2013

Accepted 6 April 2013

Available online 16 April 2013

### Keywords:

Rodents

Coprolites

Paleoparasitology

Anoplocephalids

Patagonia

## ABSTRACT

Results of paleoparasitological examination of rodent coprolites collected from archaeological and paleontological sites from Patagonia, Argentina, are present. Each coprolite was processed, rehydrated, homogenized, spontaneously sedimented and examined using light microscope. Coprolites and eggs were described, measured and photographed, and were compared with current faeces of *Lagidium viscacia*. Eggs with morphological features, attributed to an anoplocephalid cestode were found in samples collected from Cueva Huenul 1 (36°56'45"S, 69°47'32"W, Neuquén Province, Holocene) and Los Altares Profile (43°53'35"S, 68°23'21"W, Chubut Province, Late Holocene). These are the first findings of this anoplocephalid from faecal material from patagonic rodents.

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## 1. Introduction

During the last years in Argentina, paleoparasitology was mainly focussed on the study of parasites collected from archaeological sites of Patagonia. Efforts to identify parasitic remains from several fossil materials (coprolites, skeletal sediments, pellets of regurgitation, among others) were conducted, mainly focused on enteroparasites of humans, camelids, predatory birds, and micromammals ([1–5], among others). Rodents are important components of biodiversity and as hosts of numerous parasites, including those of zoonotic importance [6].

Cestodes are a ubiquitous group of intestinal and tisular parasites of all vertebrates, and are currently found in small mammals [6]. The family Anoplocephalidae (Cyclophyllidae) includes parasites infecting both terrestrial mammals (placentals and marsupials) and birds. Based on the number of genera present in these hosts, the important radiation of the anoplocephalines has taken place in rodents and lagomorphs [7,8]. Intermediate hosts are oribatid mites ingested by their herbivorous

definitive hosts [7]. Anoplocephalids are parasites of zoonotic importance for animals and humans [9,10].

Anoplocephalids have been reported from mammals from all major zoogeographic regions; however are not commonly found from Central and South America (except for species of *Monoecocestus*). According to Gardner and Campbell [11], the relative dearth of species of cestodes reported and described from South American mammals, is probably due to inadequate sampling.

Antecedents of paleoparasitological studies carried out in rodent coprolites from Patagonia revealed the presence of eggs of cestodes commonly found in micromammals, such as *Monoecocestus* spp. and *Viscachataenia quadrata* (Anoplocephalidae) [1–5].

The aim of the present study was to report the finding of eggs of anoplocephalids found in rodent coprolites collected from archaeological and paleontological sites from Patagonia (Argentina).

## 2. Materials and methods

Recently, new paleoparasitological research was started at the so-called Los Altares Profile (LAP) and Cueva Huenul 1 (CH1). Both sites are situated in Patagonia, Argentina (Fig. 1).

Cueva Huenul 1 (CH1) (36°56'45"S, 69°47'32"W) is a large archaeological cave located east of the Andes, close to the southern margin of the

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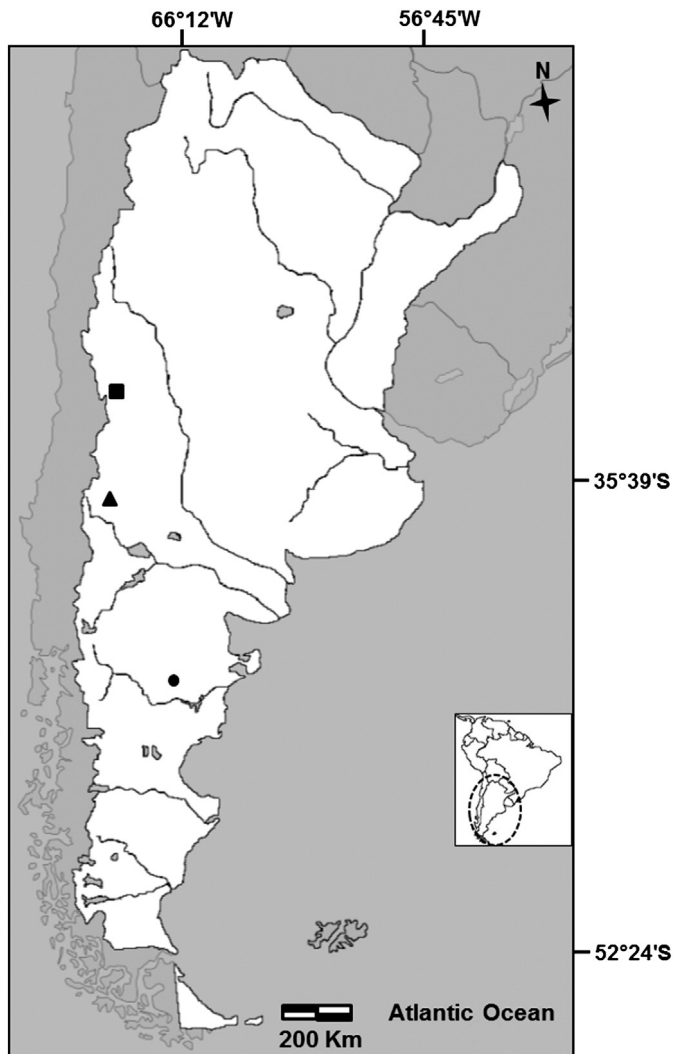


Fig. 1. Map showing the study sites: Cueva Huenul 1 (triangle), Los Altares Profile (circle) and Cajón de los Arenales (quadrangle).

Colorado River, in northern Neuquén Province. This site provides a stratified sedimentary sequence ranging from the Late Pleistocene to the Late Holocene, where coprolites found were very well preserved. Excavations provided a 1.4 m sequence composed of two sets of litho-stratigraphic units. The basal units VIII–V have a high content of organic matter, are composed mainly of megafauna dung remains, and are bracketed between radiocarbon ages of  $13,844 \pm 75$  and  $11,841 \pm 56$  yr. B.P. The second stratigraphic set (units IV–I) exhibits lower abundance of organic matter, with predominant aeolian sedimentation, and is dated between  $9531 \pm 39$  and  $1416 \pm 37$  yr. B.P. [12,13]. The site presents evidences that indicate a very brief but redundant human use of the cave [12] in different stages of the human peopling of northern Patagonia.

Los Altares Profile (LAP) ( $43^{\circ}53'2''S$ ,  $68^{\circ}23'3''W$ ) is located on the south shoulder of the National Route No. 25, 1.5 km southeast of Los Altares, Chubut Province; it corresponds to an accumulation of sedimentary fill remnant of an ancient cave eliminated by road works [14]. The sequence of 120 cm thick is formed primarily by aeolian sandy silt with variable clastic material from the weathering of country rock, layers of vegetal debris and fragments of coal. It was excavated by 12 artificial levels of 10 cm thick each. Radiocarbon dates on charcoal provided ages for the basal level (artificial level 12),  $2210 \pm 70$  yr B.P. and an intermediate level (artificial level 5),  $1280 \pm 90$  yr B.P. [14].

Forty coprolites from levels II, III, V, VI and VII from CH1 and 60 coprolites of rodents proceedings from levels I, II, IV and XII from LAP were examined for parasites. Coprolites were inventoried and processed

individually [3]. Additionally, at the end of this research, current faeces ( $N = 4$ ) of *Lagidium viscacia* collected from “Cajón de los Arenales” (CA), Mendoza province ( $33^{\circ}37'29''S$ ,  $69^{\circ}30'54''W$ ) (Fig. 1), close to CH1, were examined for parasites.

The external examination of faeces was conducted according to [15] and [16]. Each coprolite was fully processed by rehydration in a 0.5% water solution of trisodium phosphate (TSP) in a glass tube for a week, followed by homogenization, processed by spontaneous sedimentation [17] and finally preserved in ethanol 70%. Ten slides were prepared from each coprolite, along with the addition of one drop of glycerin to each slide, and examined using light microscopy. Eggs of parasites were measured and photographed at  $40\times$  magnification.

### 3. Results

Coprolites from CH1 and LAP were dark brown, concave to conical, and had a smooth surface, with one extremely dull and the other sharp. Average measurements of faeces from CH1 were  $13.62 \pm 1.84$  mm long by  $4.3 \pm 0.29$  mm wide ( $N = 5$ ); the average weight was  $0.11 \pm 0.018$  g. Average measurements of coprolites from LAP were  $11.93 \pm 1.18$  mm long by  $4.6 \pm 0.40$  mm wide, with an the average weight of  $0.09 \pm 0.018$  g ( $N = 8$ , sample 681, level I);  $13.03 \pm 1.51$  mm long by  $4.91 \pm 0.58$  mm wide, with an average weight of  $0.12 \pm 0.03$  g ( $N = 8$ , sample 591, level II); and  $15.65 \pm 1.27$  mm long by  $4.85 \pm 0.41$  mm wide, with an average weight of  $0.14 \pm 0.02$  g ( $N = 8$ , sample 593, level III).

Three of the 5 coprolites examined from level III of CH1 (Fig. 2a) and one of the 16 coprolites examined from levels I (Fig. 2b), II (Fig. 2c) and III (Fig. 2d) from LAP contained eggs of anoplocephalids (Cestoda: Anoplocephalidae) ( $N = 12$  from CH1 and  $N = 43$  from LAP), of similar morphology with features attributable to genus *Monoecocestus* Beddard, 1914 or to genus *Andrya* Railliet, 1893.

Eggs are shown as square to subrounded, because their edges are slightly folded. The embryophore presents a form of a pyriform apparatus, blunt or with short horns (Fig. 3). The average measurements of eggs from CH1 were  $72.08 \pm 6.97$   $\mu$ m long (60 to 77.5  $\mu$ m) by  $66.67 \pm 5.40$   $\mu$ m wide (60 to 75  $\mu$ m) ( $N = 6$ ); and those from LAP were  $78.20 \pm 5.84$   $\mu$ m long (67.5 to 87  $\mu$ m) by  $76.00 \pm 6.96$   $\mu$ m wide (62.5 to 87  $\mu$ m) ( $N = 25$ ).

Coprolites recovered from both sites were similar and assigned to *L. viscacia* Molina 1782 (Caviomorpha: Chinchillidae), the chinchillón or vizcacha serrana. The examination of current faeces collected from *L. viscacia* confirmed the zoological origin of the coprolites, assigned to the chinchillón, and harbored the similar anoplocephalid eggs mentioned below. The average measurements of eggs from CA were  $87.61 \pm 4.02$   $\mu$ m long (82.5 to 95  $\mu$ m) by  $75.87 \pm 5.41$   $\mu$ m wide (65 to 87.5  $\mu$ m) ( $N = 23$ ).

### 4. Discussion

The coprolites examined from both sites (CH1 and LAP) and the current faeces from CA were similar in aspect. Based on the morphology and size of the faecal material, and on the coproparasitological knowledge on similar coprolites [1], the samples were attributed to *L. viscacia* Molina 1782 (Caviomorpha: Chinchillidae), the chinchillón or vizcacha serrana. The family Chinchillidae contains chinchillas, viscachas and their fossil relatives. The family is restricted to southern and western South America [18,19].

The families Cricetidae, Ctenomyidae, Caviidae and Didelphidae dominate small mammal samples at CH1, recovered from Late Pleistocene to Late Holocene layers, and at LAP, from the Late Holocene. In both sites, the absence of cut-marks, presence of light digestive marks, presence of few burned remains, and low abundance of some large (> 200 g), mostly diurnal, gregarious or colonial rodents, are indicative of non-human deposition. These accumulations would be mainly due to the feeding activity of the Common Barn Owl *Tyto alba* [13,14,20]. The



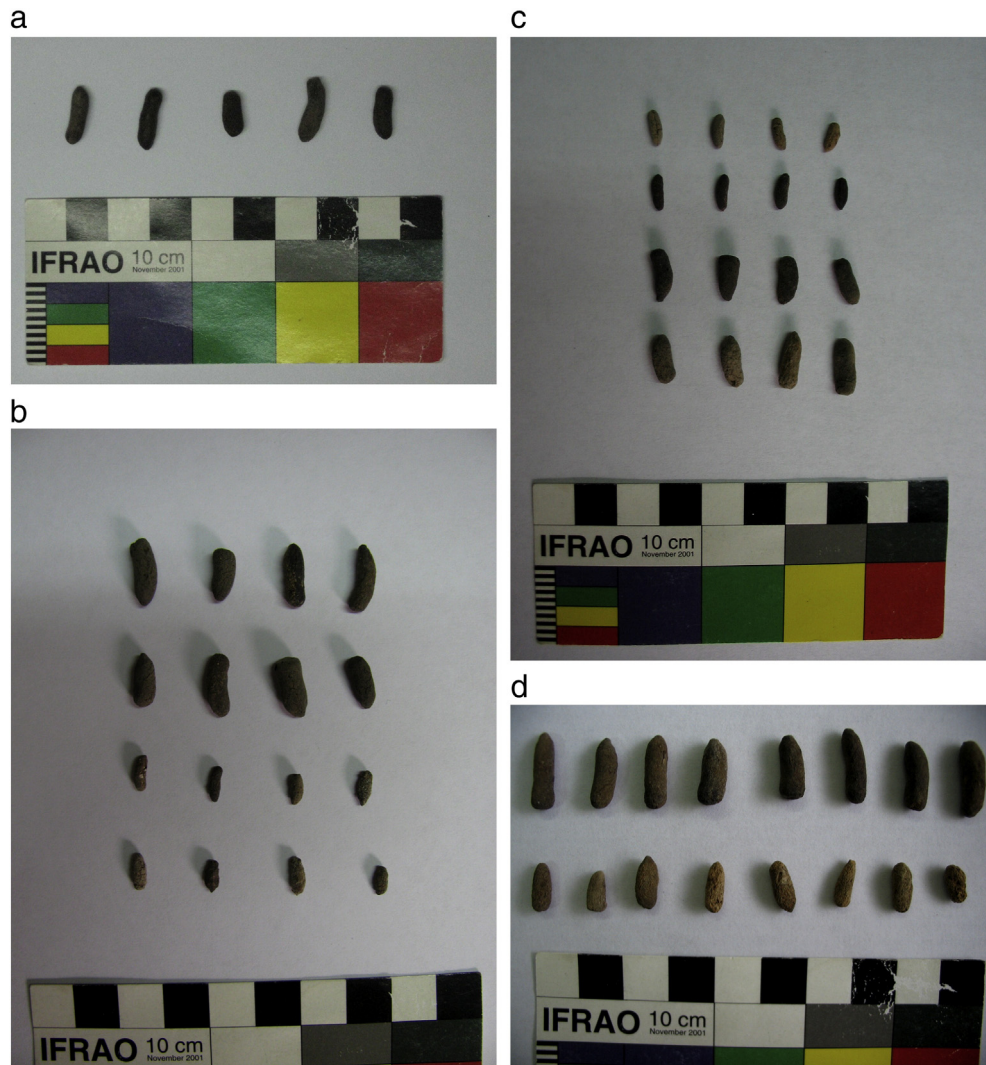


Fig. 2. Macroscopic aspect of the coprolites examined. Coprolites from Cueva Huenul 1 (a), Los Altares Profile level I (b), level II (c) and level III (d).

poor bone quantity of *L. viscaciae* in the small mammal's records is probably due to this type of archaeofaunistic owl pellet accumulations, as owls do not feed on large rodents.

Anoplocephalids (Cyclophyllidae) are very well represented in small mammals with 25 genera described at present [8]. Fewer than 30 species of anoplocephaline cestodes (mostly *Monoecocostus* spp.) have been described at present from mammals in the Neotropics,



Fig. 3. Anoplocephalid (Cyclophyllidae: Anoplocephalidae) eggs observed in ancient samples. Bar = 20  $\mu$ m.

and all of them were found in hystricognath and sigmodontine rodents [21]. The only known valid anoplocephalid genera of South American rodents are *Monoecocostus*, *Andrya* and *Viscachataenia* (Global Cestode Database).

Anoplocephalids known at present collected from South American rodents according to the bibliography searched and the Global Cestode Database are: *Monoecocostus myopotami* in *Myocastor coypus* from Argentina [22]; *M. andersoni* and *M. microcephalus* in *Graomys domorum*, *M. eljeje* and *M. petiso* in *Galea musteloides*, *M. poralus* in *Phyllotis caprinus*, *M. sininterus* in *P. wolffsohni*, and *M. threlkeldi* in *Holochilus brasiliensis*, all of which are from Bolivia [21]. *M. threlkeldi* in *Lagidium peruanum* is from Perú [23]. *Monoecocostus* spp. observed in rodents from Brazil are *M. hagmanni* [23], *M. jacobi* [24] and *M. macrobursatum* [25] in *Hydrochoerus hydrochaeris*, *M. machadoi* in *Proechymis guyannensis* [7], *M. minor* in *Cavia aperea* [26] and *M. parcitesticulatus* in *Cavia porcellus* [27]. From Paraguay, *M. hydrochoeri* in *H. hydrochaeris* and *M. mackiewiczii* in *Phyllotis* sp. were reported [28]. Finally, from Chile there is a record of *M. torresi* in *Ctenomys maulinus* [29].

Hystricognath rodents are the dominant host for species of *Monoecocostus*, but Haverkost and Gardner [21] indicate that the sigmodontine rodents (Myomorpha: Cricetidae: Sigmodontinae) are suitable hosts for these helminths as well.

In relation to *Andrya*, records from South American rodents are *Andrya octodonensis*, reported in *Phyllotis xanthopygus* from Argentina [30] and in *Octodon degus* from Chile [31]. *A. vesicula* n. sp. was reported

in *Phyllotis xanthopygus* and *A. boliviensis* was registered in *Phyllotis osliae* from Bolivia [30].

*Viscachataenia quadrata* was found in *L. viscacia* [32] from Argentina and in *L. peruanum* from Perú [33].

Previous paleoparasitological studies on archaeological sites revealed the presence of cestode eggs in rodents. Eggs of *Monoecocestus* sp. were found in rodent coprolites from Alero Mazquiarán (Chubut province), assigned to the interface of the Araucanian and Tehuelche cultures, dated at  $212 \pm 35$  years B.P. [4] and from Alero Destacamento Guardaparque, located in the Perito Moreno National Park (Santa Cruz province) from Middle Holocene levels [5]. Beltrame et al. [1] found eggs attributable to *Viscachataenia quadrata* and *Monoecocestus* sp. in coprolites probably of *L. viscacia* from CH1 dated at the Late Pleistocene/Early Holocene transition to the Late Holocene period. Anoplocephalid eggs were also recorded from a rockshelter close to Río Mayo locality, southwest of Chubut province associated to European contact, probably the XIX Century [2,3].

Anoplocephalid species are very difficult to identify by their eggs. There are some interspecific size differences, but these could be affected by methods of preservation. In this sense, the eggs found in the present study, based on their aspect and size, could be attributed either to *Monoecocestus* or *Andrya*. *Viscachataenia* was not considered because its eggs are four-lobed. The differences in the measurements of the eggs found between current and ancient samples can be attributed to preservation methods or taphonomic processes.

Uterine morphology has played a key role in the systematic and phylogenetic arrangements within anoplocephaline cestodes [8]. Eggs are not taken into account at present for taxonomic differences among genera. Nevertheless, the importance of the study of anoplocephalid eggs is evident, mainly in paleoparasitological and environmental studies, since eggs are the parasitic remains most commonly found. It is necessary to improve their published descriptions and illustrations for future taxonomic studies.

Oribatid mites are intermediate hosts for anoplocephalids, and are commonly ingested by herbivorous where infection occurs. Anoplocephalids can cause human disease if humans eat mites present in the soil [9]. Humans living in CH1 were probably exposed to illness by these cestodes during the entire period of time considered.

## Acknowledgements

This work was supported by Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT): PICT 2010–1856, PICT 2010–2665 and CREOI.

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