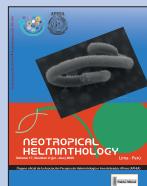


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## ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

### CONTRIBUTION TO THE KNOWLEDGE OF PARASITIC NEMATODES IN MAMMALS FROM SAN JUAN, ARGENTINA

### CONTRIBUCIÓN AL CONOCIMIENTO DE NEMÁTODOS PARÁSITOS EN MAMÍFEROS DE LA PROVINCIA DE SAN JUAN, ARGENTINA

Cynthia J. González-Rivas<sup>1,2</sup>, Gabriel N. Castillo<sup>2,3,4\*</sup> & Ivan D. Simoncelli<sup>1</sup>

<sup>1</sup> Faunístico: Centro de Rehabilitación de Fauna Silvestre, Educación Ambiental y Recreación Responsable, San Juan, Argentina. Ruta Provincial N° 60 km 14 5400 Rivadavia, San Juan, Argentina.

<sup>2</sup> Parasitología en animales silvestres. Departamento de Biología, Facultad de Ciencias Exactas Físicas y Naturales. Universidad Nacional de San Juan. Av. Ignacio de la Roza 590, 5402, San Juan, Argentina.

<sup>3</sup> Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET). CIGEOBIO (Centro de Investigaciones de la Geósfera y Biósfera) CONICET-UNSJ, Av. Ignacio de la Roza 590, 5402, San Juan, Argentina.

<sup>4</sup> Gabinete de Investigación Diversidad y Biología de Vertebrados del Árido (DIBIOVA). Universidad Nacional de San Juan. Av. Ignacio de la Roza 590, 5402, San Jun, Argentina.

\* Corresponding author: liolaemusparvus@gmail.com

Cynthia J. González-Rivas: <https://orcid.org/0000-0001-8294-6999>

Gabriel N. Castillo: <https://orcid.org/0003-1519-9457>

Ivan Dario Simnocelli: <https://orcid.org/0009-0002-1251-6293>

## ABSTRACT

The endoparasitic fauna of four mammal species was analyzed: *Puma concolor* (Linnaeus, 1771) (cougar), *Lycalopex gymnocercus* (Fischer, 1814) (grey fox), *Myocastor coypus* (Molina 1782) (coipus) and *Tolypeutes matacus* (Linnaeus, 1758) (armadillo). These specimens entered the Center for Wildlife Rehabilitation, Environmental Education, and Responsible Recreation located in the province of San Juan, Argentina. Out of the four mammal species, three died on the spot. The necropsy of *L. gymnocercus*, *M. coypus*, and *T. matacus* was carried out, and it was evidenced the presence of three species of parasitic nematodes: *Physaloptera clausa* Rudolphi, 1819 (Physalopteridae) in *L. gymnocercus*; *Trichuris myocastoris* Enigk, 1933 (Trichuridae) in *M. coypus*; and adult females of *Aspidodera* sp. Railliet & Henry, 1912 in *T. matacus*. Regarding *P. concolor*, the expulsion of nematodes was done orally, determining the presence of *Toxascaris* sp. Linstow, 1902 larvae (Toxocaridae). Metric and morphological data of the evidenced nematodes are presented. The present work mentions four new parasite-host interactions for Argentina, and the first records for the province of San Juan, thus contributing to the knowledge of parasites in mammals.

**Keywords:** *Lycalopex gymnocercus* – *Myocastor coypus* – nematodes – *Puma concolor* – San Juan – *Tolypeutes matacus*

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## RESUMEN

Analizamos la fauna endoparasitaria de cuatro especies de mamíferos; *Puma concolor* (Linnaeus, 1771) (puma), *Lycalopex gymnocercus* (Fischer, 1814) (zorro gris), *Myocastor coypus* (Molina, 1782) (coipus) y *Tolypeutes matacus* (Linnaeus, 1758) (armadillo de tres bandas) del Centro de Rehabilitación de Vida Silvestre, Educación Ambiental y Recreación Responsable en la provincia de San Juan, Argentina. De las cuatro especies de mamíferos que ingresaron al Centro de Rehabilitación, tres fallecieron en el lugar, posteriormente se procedió a realizar la necropsia. El análisis post mortem de *L. gymnocercus*, *M. coypus* y *T. matacus* evidencio la presencia de tres especies de nemátodos parásitos; *Physaloptera clausa* Rudolphi, 1819 (Physalopteridae) en *L. gymnocercus*, *Trichuris myocastoris* Enigk, 1933 (Trichuridae) en *M. coypus*, y hembras adultas de *Aspidodera* sp. Railliet & Henry, 1912 en *T. matacus*. Con respecto al *P. concolor*, expulsó los nemátodos por vía oral, determinándose la presencia de larvas de *Toxascaris* sp. Linstow, 190 (Toxocaridae). Se presentan datos métricos y morfológicos de los nemátodos. Nuestro trabajo menciona cuatro nuevas interacciones parásitos- hospedadores para Argentina y los primeros registros para la provincia de San Juan, contribuyendo de esta forma al conocimiento parasitario en mamíferos.

**Palabras claves:** *Lycalopex gymnocercus* – *Myocastor coypus* – nematodos – *Puma concolor* – San Juan – *Tolypeutes matacus*

## INTRODUCTION

The Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunístico) is the only institution for the conservation of wildlife in the province of San Juan, Argentina. Some of the objectives of such institution are environmental education, rehabilitation of species and research. Among the research tasks is to obtain knowledge of the health status of the specimens that enter the Wildlife Rehabilitation Center, which is why parasitological analyzes are of primary concern to take health measures mainly in mammalian species (González-Rivas *et al.*, 2021).

Currently, the knowledge of parasites of wild mammals in the province of San Juan is extremely scarce and unknown. Approximately 50 species of mammals have been mentioned for the province of San Juan (Bauni *et al.*, 2021). Of these, only three species of mammals present endoparasitic records: *Lama guanicoe* Müller, 1776 (Artiodactyla, Camelidae); *Chaetophractus vellerosus* Gray, 1865 (Cingulata, Chlamyphoridae); and *Phyllotis xanthopygus* Waterhouse, 1837 (Rodentia, Cricetidae) (Ezquiaga, 2013; Castillo *et al.*, 2016; González-Rivas *et al.*, 2019). The rest of the mammalian species have not been studied from an endoparasitic point of view.

Due to the lack of parasitic knowledge and studies carried out in mammals from San Juan, we examined four species of mammals: *Puma concolor* (Linnaeus, 1771), *Lycalopex gymnocercus* (Fischer, 1814), *Myocastor coypus* (Molina, 1782) and *Tolypeutes matacus* (Linnaeus, 1758) that entered the Rehabilitation Center, San Juan province, Argentina.

## MATERIALS AND METHODS

The information presented in the following study was based on the recognition of endoparasites obtained from four species of mammals belonging to the Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunístico), situated in the department of Rivadavia, San Juan province, Argentina. The species analyzed came from the phytogeographic region of Puna and Monte, province of San Juan, Argentina. The Puna region is characterized by a cold and dry climate, with great thermal amplitude, strong winds, high radiation and prolonged periods of drought (Márquez *et al.*, 2016). The characteristic woody vegetation is represented by thickets of pinchaguas (*Lycium chinar* Phil, 1891; *L. fuscum* Miers, 1854), wormwood (*Artemisia mendozana* var. *paramilloensis* Roig & Ambrosetti 1989), tolas (*Fabiana denudata* Miers, 1846), yellow firewood (*Adesmia horrida* Gillies, Hook & Arn, 1832), pingopingo (*Ephedra multiflora* Phil, 1889), among others. The herbaceous stratum presents several gramineous species of the genus *Stipa* and *Jarava* (Márquez *et al.*, 2016). The Monte region covers extensive arid areas with an average rainfall of less than 100 mm/year. It has an area of 40,499 km<sup>2</sup>, corresponding to 45% of the total of the province of San Juan. Xerophytic plants adapted to a hot and dry climate predominate. The vegetations that predominate are Zygophyllaceae, Malpighiaceae and Fabaceae. Among the family Zygophyllaceae we can mention jarilla (*Larrea divaricata* Cav 1800, *Larrea cuneifolia* Cav 1800), retamo (*Bulnesia retama* Gillies ex Hook & Arn 1874), manca potrillo (*Plectrocarpa tetracantha* Gillies ex Hook & Arn 1833); from Malpighiaceae we can mention *Tricomaria*

*ucillo* Hook & Arn 1833; and from Fabaceae there are shrubby species such as *Prosopis*, the tintitaco (*Prosopis torquata* Cav. ex Lag. 1825), lamar (*Prosopis alpataco* Phil 1862) (Márquez *et al.*, 2016).

Between January 2019 and November 2020 the following species were analyzed: *Puma concolor* (Felidae) (n=1) from the Puna ecoregion; and *Lycalopex gymnocercus* (Canidae) (n=1), *Myocastor coypus* (Echimyidae) (n=1) and *Tolypeutes matacus* from the Monte ecoregion, San Juan province, Argentina.

Three of the mammalian species (*M. coypus*, *L. gymnocercus* and *T. matacus*) entered in poor sanitary conditions, which subsequently died on the spot. During post-mortem examinations, parasitic nematodes were identified in stomachs and intestines.

In the case of *P. concolor*, the nematodes were obtained by regurgitation (expulsion from the stomach). The parasites were collected and preserved in 70° ethyl alcohol. For observation, identification/determination of nematodes, semi-permanent preparations were made using lactophenol solution. Parasitic intensities were determined according to Bush *et al.* (1997). In all cases, the mammal species were studied in the laboratory of the Wildlife Rehabilitation Center (Faunístico).

Metric and morphological data of the nematodes are presented. Measurements are in micrometers (μm) unless otherwise indicated. Mean, standard deviation and range

are presented between parentheses. The nematodes are deposited in the Parasitological Collection of the Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunístico): CRFSJ\_P\_26, CRFSJ\_P\_36 and CRFSJ\_P\_39.

**Ethic aspects:** All applicable international, national, and/or institutional guidelines for the care and use of animals were strictly followed. All animal sample collection protocols complied with the current laws of Argentina. All animal procedures performed in this research were in accordance with the ethical standards of the institution or organization at which the study was conducted.

## RESULTS

Four species of parasitic nematodes were identified: *Toxascaris* sp. (Toxocaridae), *Physaloptera clausa* (Physalopteridae), *Trichuris myocastoris* Enigk, 1933 (Trichuridae) y *Aspidodera* sp. (Heterakoidea).

Order Carnivora

Family Felidae

*Puma concolor* (Linnaeus, 1771) (Fig. 1)

**Intensity:** 2 larvae of *Toxascaris* sp.

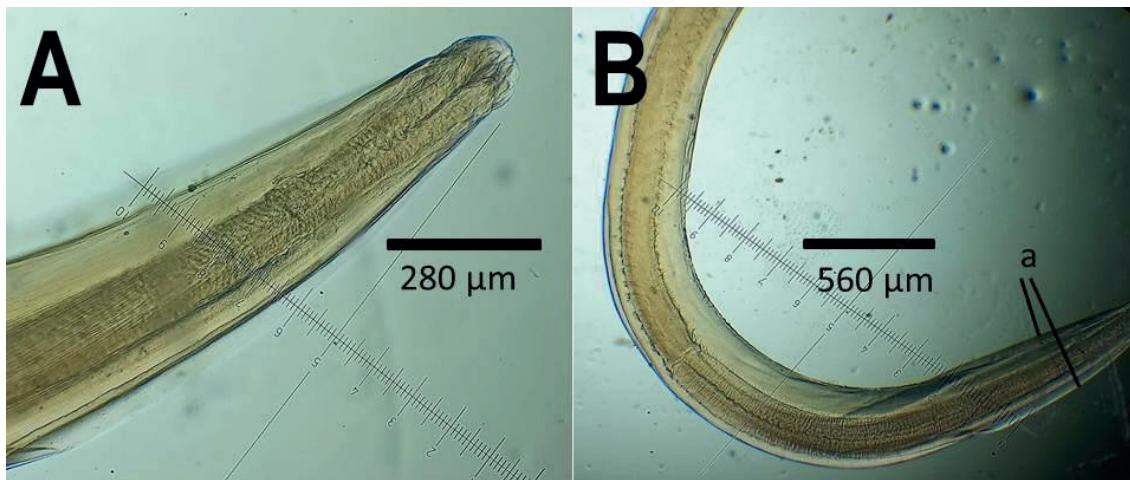
*Toxascaris* sp. Linstow, 1902 (Spirurida Ascarididae)  
(Fig. 2)



**Figure 1.** Specimen of *Puma concolor* (Photo: Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation).

**General:** Large and white nematodes. Intestinal caecum absent. Interlips absent. Cervical wings are slender and lance-shaped.

**Larvae (based on 2 specimens) ( $\mu\text{m}$ ):** Length (mm)  $11.5 \pm 0.7$  (11–12); total width  $286 \pm 4.9$  (283–290); cervical wing length  $1137 \pm 3.5$  (1135–1140); cervical wing width  $44 \pm 1.4$  (43–45); distance of nerve ring 485  $\pm 7.07$  (480–490); muscular esophagus length 2925  $\pm 36$  (2900–2951).



**Figure 2.** Larval stage *Toxascaris* sp. recorded in *P. concolor*. Anterior view (A); middle and anterior view (B). The lance-shaped cervicals are observed= a.

Order Carnivora

Family Canidae

*Lycalopex gymnocercus* (Fischer, 1814) (Fig. 3)

**Intensity:** 3 males and 7 females of *Physaloptera clausa*

*Physaloptera clausa* Rudolphi, 1819 (Spirurida: Physalopteridae) (Fig. 4)

**General:** White nematodes. Cuticle thick, with transverse striations. Evident sexual dimorphism. Cephalic collar present. Oral opening surrounded by two lateral pseudolabia; each pseudolip with two cephalic papillae. Esophagus with a short muscular and long glandular portion. The nerve ring surrounds the muscular esophagus. Spicules of different size, unequal in length; left spicule longer and narrower; short right spicule, wider than left spicule. 4 pairs of pedunculated papillae. Anterior cloaca region with 3 sessile papillae (median papilla larger than lateral papillae), posterior margin with 2 pairs of sessile papillae. 3 additional pairs of sessile postcloacal ventral papillae.

**Site of infection:** Stomach.

**Collection place:** Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunistic).

**Deposited material:** CRFSJ\_P\_36.

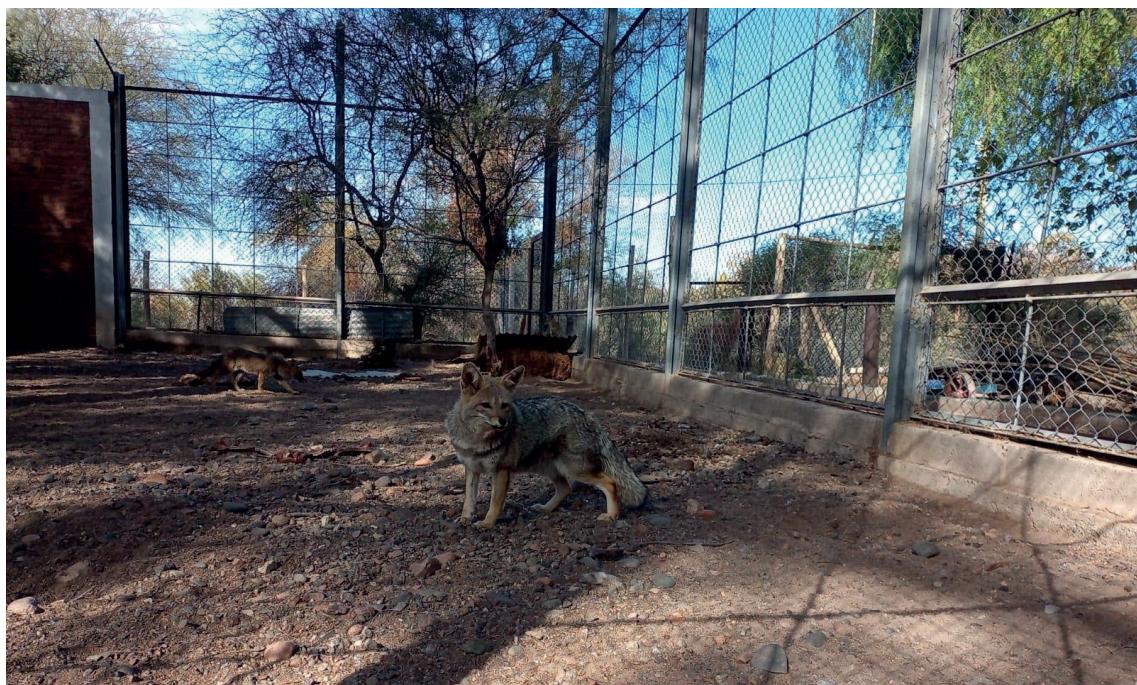
**Male (based on 3 adult specimens) ( $\mu\text{m}$ ):** Total length (mm)  $35.5 \pm 1.86$  (34–37.6); width at level of excretory pore  $838 \pm 1$  (837–839); muscular esophagus length  $685 \pm 4$  (681–689); glandular esophagus length  $2498.6 \pm 1.53$  (2497–2500); nerve ring  $454.6 \pm 1.15$  (454–456); right spicule  $438 \pm 17.09$  (420–454); left spicule  $796.3 \pm 2.08$  (794–798).

**Female (based on 2 gravid adults) ( $\mu\text{m}$ ):** Total length (mm)  $45.5 \pm 2.12$  (44–47); width at level of excretory pore  $1032 \pm 4.24$  (1029–1035); muscular esophagus length  $685.5 \pm 6.36$  (681–690); glandular esophagus length  $4520 \pm 28.2$  (4500–4540); nerve ring  $558.5 \pm 12$  (550–567); vulva distance (mm)  $7.5 \pm 0.71$  (7–8); egg width  $25.5 \pm 0.7$  (25–26); egg length  $37.7 \pm 0.35$  (37.5–38).

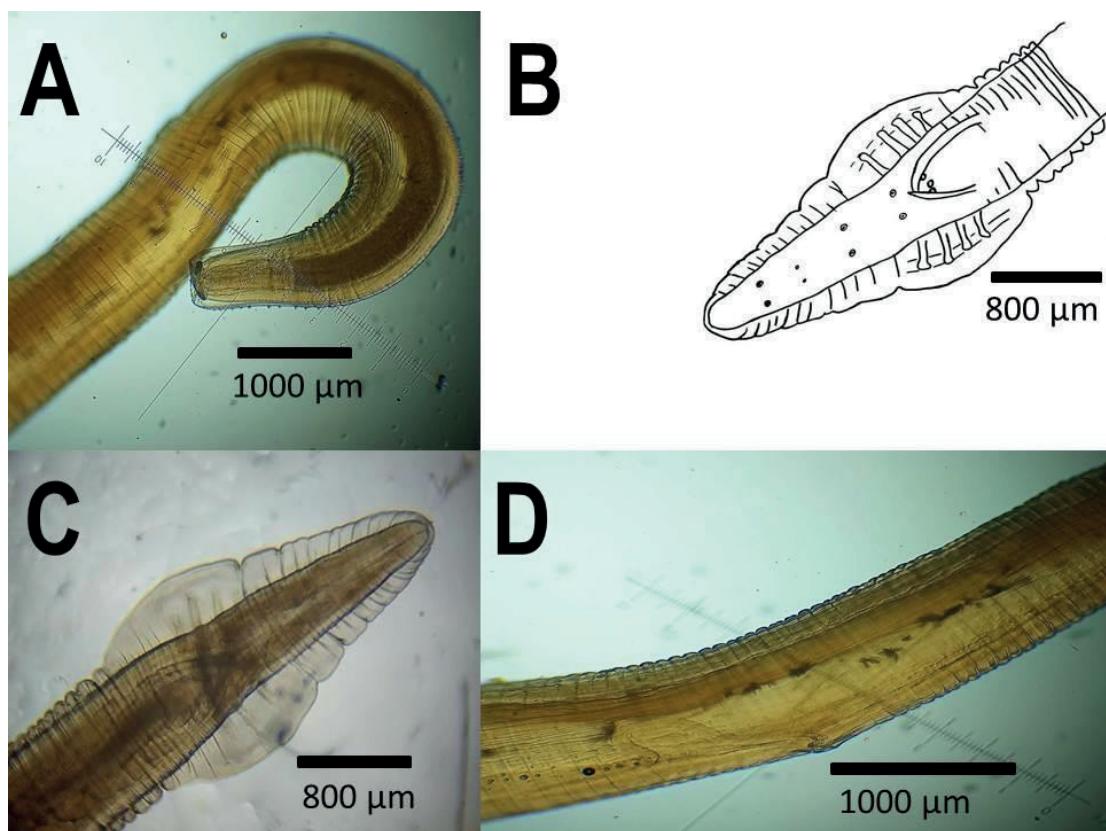
**Site of infection:** Stomach.

**Collection place:** Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunistic).

**Deposited material:** CRFSJ\_P\_26.



**Figure 3.** Specimen of *Lycalopex gymnocercus* (Photo: Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation).



**Figure 4.** *Physaloptera clausa* recorded on *L. gymnocercus*. Anterior view, male (A); posterior view, male (B and C); middle view, female (D).

Order Rodentia

Family Echimyidae

*Myocastor coypus* (Molina 1782) (Fig. 5)

**Intensity:** 5 males and 8 females of *Trichuris myocastoris*

*Trichuris myocastoris* Enigk, 1933 (Trichocephalida: Trichuridae) (Fig. 6, 7)

**General:** Robust, white, non-fliform body. Transversely striated cuticle. Males with spiral back of body; spicule in a foreskin-like sheath, with spiny outer surface. Females with slightly curved posterior end, not spiral. Vulva near the junction of the two body parts. Oviparous. Lemon-like eggs with a thick brown peel and a polar thickening at each end.

**Male (based on 2 adult specimens) (μm):** Total length (mm)  $42.5 \pm 0.7$  (42–43); posterior width  $391 \pm 1.4$

(390–392); anterior length (mm)  $27.5 \pm 0.7$  (27–28); anterior width  $57 \pm 1.4$  (56–58); spicule length (mm)  $2.1 \pm 0.1$  (2–2.20); spicule width  $29.5 \pm 0.7$  (29–30).

**Female (based on 2 gravid adult specimens) (μm):** Total length (mm)  $44.5 \pm 2.1$  (43–46); posterior width  $564 \pm 5.6$  (560–568); anterior length (mm)  $23 \pm 1.4$  (22–24); anterior width  $133 \pm 4.2$  (130–136); vulva distance (mm)  $26 \pm 1.4$  (25–27); wide egg  $39.5 \pm 0.7$  (39–40); egg length  $66.5 \pm 2.12$  (65–68).

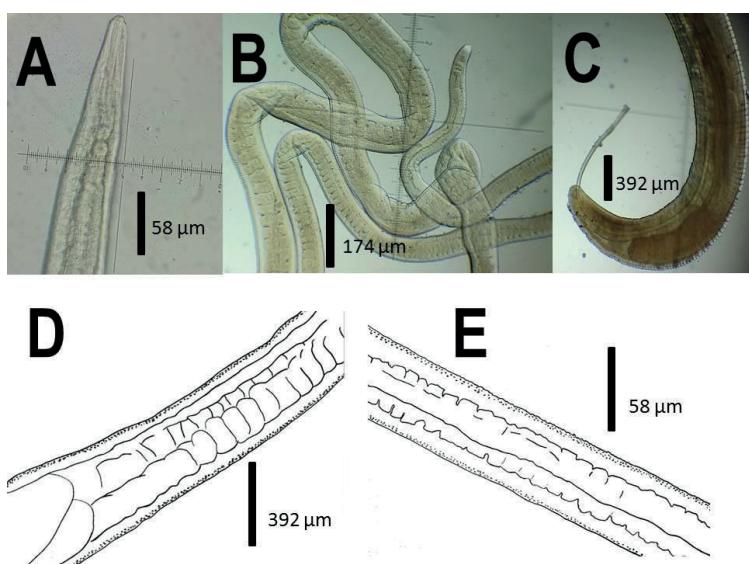
**Site of infection:** Large intestine and cecum.

**Collection place:** Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Faunistic).

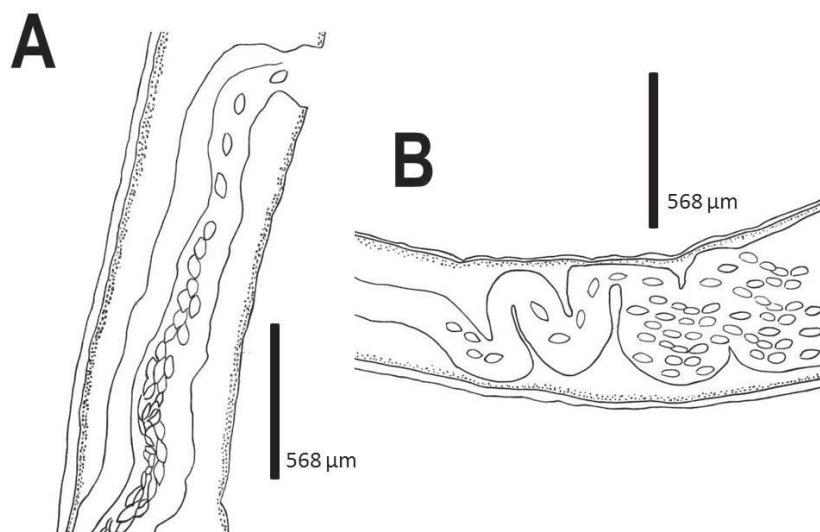
**Deposited material:** CRFSJ\_P\_36.



**Figure 5.** Specimen of *Myocastor coypus* (Photo: Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation).



**Figure 6.** *Trichuris myocastoris* recorded on *M. coypus*, adult male. Anterior view (A and B); posterior view, spicule (C); middle view (D); detail of anterior view (E).



**Figure 7.** *Trichuris myocastoris* recorded on *M. coypus*, female, adult. Median view, with eggs and vulva (A); median view with eggs (B).

Order Cingulata  
Family Chlamyphoridae  
*Tolypeutes matacus* (Linnaeus, 1758) (Fig. 8)

**Intensity:** 5 adult gravid females of *Aspidodera* sp.

*Aspidodera* sp. Railliet & Henry, 1912 (Ascaridida: Aspidoderidae) (Figs. 9, 10)

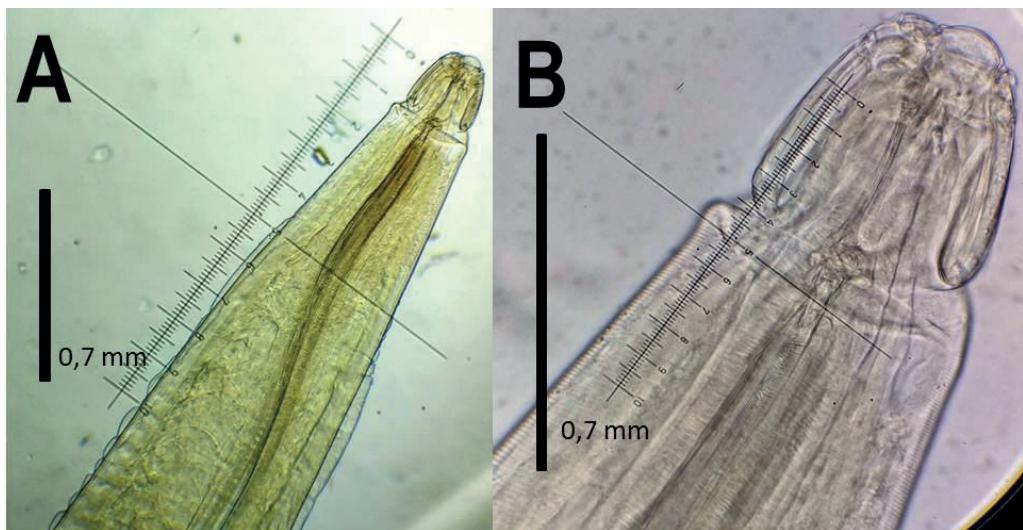
**General:** Adult nematodes with presence of recurrent and anteriorly anastomosed cephalic cords. Mouth with

three lips. Nerve ring located near the middle esophagus. Gravid females, with the vulva near the middle of the body. Eggs with ellipsoid shape, thick and smooth shell.

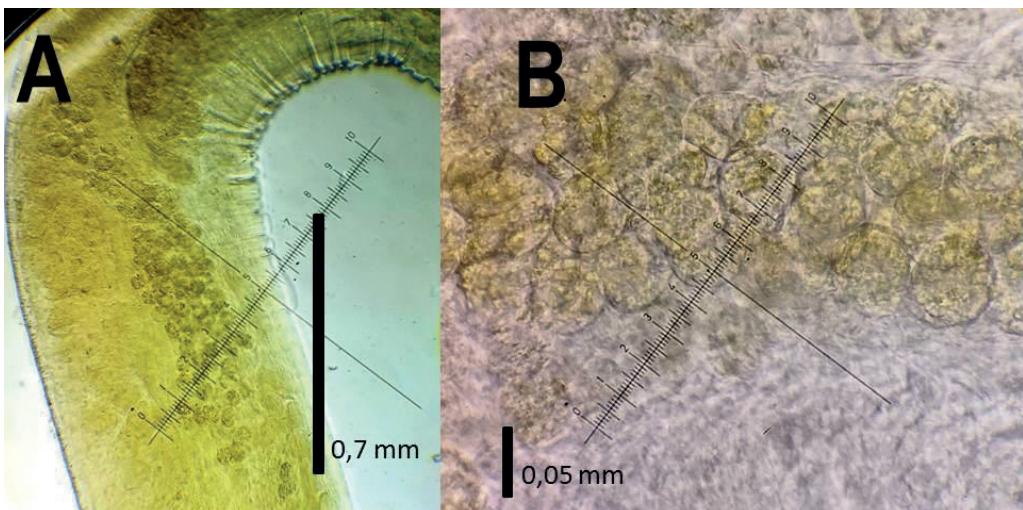
**Female (based on 2 gravid adult specimens) (mm):**  
Total length  $7.95 \pm 0.07$  (7.9– 8); total width  $0.7 \pm 0.03$  (0.68– 0.72); esophagus length (including the bulb)  $2.38 \pm 0.03$  (2.36– 2.40); Distance of nerve ring  $1.45 \pm 0.07$  (1.40– 1.50); vulva length  $4.45 \pm 0.07$  (4.40– 4.50); egg width 0,04 (0.04– 0.05); egg length 0.05 (0.05).



**Figure 8.** Specimen of *Tolypeutes matacus* (Photo: Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation).



**Figure 9.** *Aspidodera* sp. recorded on *T. matacus*, female, adult. Anterior view (A) and detail of cephalic cords (B).



**Figure 10.** *Aspidodera* sp. recorded on *T. matacus*, female, adult. Middle view (A) and eggs (B).

## DISCUSSION

We analyzed the wildlife of mammals in the province of San Juan, Argentina, in search of endoparasites. Our analyzes allowed us to identify *Toxascaris* sp. (Toxocaridae) in *P. concolor*, *P. clausa* (Physalopteridae) in *L. gymnocercus*, *T. myocastoris* (Trichuridae) in *M. coypus* and gravid adult females of *Aspidodera* sp. in *T. matacus*. All the records found turn out to be new parasite-host interactions and the first records for the province of San Juan. Only 11 taxa of endoparasites in wild mammals had been mentioned in the province of San Juan so far: coccidia of *Eimeria macusaniensis* Guerrero, Hernandez, Bazalar & Alva, 1971, *Eimeria ivitaensis* Leguiá & Casas, 1998,

*Eimeria* sp. Schneider, 1875, (Apicomplexa: Eimeriidae), *Nematodirus* sp. Ransom, 1907 (Nematodirinae: Molineidae) and *Trichuris* sp. (Linnaeus, 1771) (Trichocephalida: Trichuridae) in *Lama guanicoe* (Müller, 1776) (González-Rivas et al., 2019); *Pterygodermatites* (P) *argentinensis* Ezquiaga, Rios, Abba & Navone, 2017, *Pterygodermatites* (P) *chaetophracti* Navone & Lombrero, 1980, *Trichohelix tuberculata* (Parona & Stossich, 1901), *Aspidodera fasciata* (Schneider, 1866) and *Aspidodera scoleciformis* (Diesing, 1851) in *Chaetophractus vellerosus* (Gray, 1865) (Ezquiaga, 2013; Ezquiaga et al., 2013; 2017) and *Pterygodermatites* (P) *kozeki* has been recorded in *Phyllotis xanthopygus* (Waterhouse 1837) (Castillo et al., 2016). In our work we mention 4 new records of

parasites, as well as new interactions with hosts never mentioned before for the province of San Juan. In this way, we raise to 15 the endoparasite species mentioned in wild mammals in San Juan.

The most common species to find in *P. concolor* are *Toxocara canis* (Werner, 1782), *Toxocara cati* (Schrank, 1788), and *Toxascaris leonina*. We mention larval stages of *Toxascaris* (Leiper, 1907) in *P. concolor*, which were expelled orally. The genus *Toxascaris* is made up of 6 species (Hodda, 2022), with a monoxene cycle and veterinary importance among predatory mammals of the Felidae families (Okulewicz *et al.*, 2012; Taylor *et al.*, 2016; Fugassa, 2020). The infective stage is the egg containing 2 or 3 larvae. The eggs develop to the infective stage rapidly. After ingestion and hatching, the larvae enter the wall of the small intestine and remain for about 2 weeks. Adult stages appear approximately 6 weeks after infection (Taylor *et al.*, 2016). In *Toxascaris*, the cervical wings are long and narrow, resembling a spear, a characteristic that differentiates it from *Toxocara cati* (Schrank, 1788). However, in *T. cati* the cervical wings are short, similar to an arrowhead (Taylor *et al.*, 2016). The *Toxascaris* nematode has been recorded in *P. concolor* in different South American countries (Fugassa, 2020), with a single record in Argentina in the province of Santa Cruz (Moleón *et al.*, 2015; Vega *et al.*, 2018; Fugassa, 2020). Our work reports a new mention, this being the second record for *Toxascaris* in *P. concolor* for Argentina and the first mention for the province of San Juan.

The genus *Physaloptera* Rudolphi, 1819, is made up of a total of 144 species (Hodda, 2022) and includes stomach parasites of mammals and other vertebrates (Anderson *et al.*, 1974; Taylor *et al.*, 2016). The life cycle is heteroxenous, where eggs expelled in the feces develop into infective larvae when ingested by coprophagous beetles, crickets, and other insects. The life cycle is completed when the definitive hosts ingest intermediate hosts (Taylor *et al.*, 2016). In Argentina, there is only one mention of *P. clausa* in the fox *L. gymnocercus*, corresponding to the province of Neuquén (Stein *et al.*, 1994; Fugassa, 2020). In our work we mention the second record for Argentina in the gray fox *L. gymnocercus* and the first record for the province of San Juan.

The genus *Trichuris* contains 107 species (Hodda, 2022) with a cosmopolitan distribution, being found in the cecum and colon of various mammalian species (Taylor *et al.*, 2016; Robles & Navone, 2014). These species are known as "whipworms" due to a broad back end and a long, filamentous front end (Anderson *et al.*, 1974; Taylor *et al.*, 2016). The back end of the male is highly

coiled and it has a single spicule in a protractile sheath (Taylor *et al.*, 2016). *Trichuris myocastoris* was described by Enigm (1933), later Baruš *et al.* 1975 redescribed the species (Rylková *et al.*, 2015). This species has mainly been mentioned in *M. coypus* in different localities in South America (Fugassa, 2020), and it has been mentioned twice parasitizing *M. coypus* in Argentina, in the province of Buenos Aires and Chaco, by Martino *et al.* (2012) and Boero & Boehringer (1967). Our work reports a new mention of *T. myocastoris*, this being the third record of *T. myocastoris* in *M. coypus* for Argentina and the first for the province of San Juan.

The genus *Aspidodera* Railliet & Henry, 1912, contains mainly parasites of marsupials, Xenarthrans and rodents in South America (Anderson *et al.*, 1974), currently with 9 valid species (Hodda, 2022). The different species of *Aspidodera* have been recorded in mammals of the order cingulata, with records of *Aspidodera binansata* (Railliet & Henry, 1913), *Aspidodera fasciata* (Schneider, 1866), *Aspidodera raillieti* (Travassos, 1913), *Aspidodera scoleciformis* (Diesing, 1851) and *Aspidodera vazi* (Proença, 1937) in *Chlamyphorus truncatus* Harlan, 1825, *Chaetophractus villosus* (Desmarest, 1804), *Chaetophractus vellerosus*, *Dasypus hybridus* (Desmarest, 1804) and *T. matacus* (Fugassa, 2020). In San Juan, only *A. fasciata* and *A. scoleciformis* have been mentioned in *Ch. vellerosus* for the Usno locality, department of Valle Fétil. We mention the genus *Aspidodera* sp. in the armadillo *T. matacus*.

Our work contributes to the parasitic knowledge of wild mammals by mentioning new parasite-host interactions for the province of San Juan and Argentina.

#### **Author contributions: CRediT (Contributor Roles Taxonomy)**

**CJGR** = Cynthia J. González-Rivas

**GNC** = Gabriel N. Castillo

**IDS** = Ivan D. Simoncelli

**Conceptualization:** CJGR, GNC, IDS

**Data curation:** CJGR, GNC

**Formal Analysis:** GNC

**Funding acquisition:** CJGR, IDS

**Investigation:** CJGR, GNC

**Methodology:** GNC

**Project administration:** CJGR

**Resources:** IDS

**Software:** CJGR, GNC**Supervision:** GNC**Validation:** CJGR, GNC**Visualization:** CJGR, GNC**Writing – original draft:** GNC**Writing – review & editing:** CJGR

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