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NEW RECORDS OF HELMINTH PARASITES IN AMPHIBIANS FROM THE SOUTH AMERICAN CHACO

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New Records of Helminth Parasites in Amphibians from the South American Chaco. González, M. A., Villegas Ojeda, M. A., Caraballo, J. N., Ailán Choke, L. G., Hamann, M. I., Duré, M. I., Gómez, V. I. & González, C. E. — This study aims to identify the helminth fauna of anurans from Argentina's Dry and Humid Chaco ecoregions. We examined 152 specimens of 12 anuran species belonging to the families Hylidae, Leptodactylidae, and Microhylidae from 3 areas located in the provinces of Chaco and Formosa (Dry Chaco) and in Corrientes province (Humid Chaco). A total of 17 helminth taxa were found: acanthocephalans in the larval stage (*Centrorhynchus* sp.), nematodes in the larval stage (*Contraecaecum* sp., *Porrocaecum* sp., *Physaloptera* sp., aff. *Cystidicolidae*), and nematodes in the adult stage (*Rhabdias* sp., *R. elegans*, *Strongyloides pereirai*, *Oswaldocruzia proencai*, *O. subauricularis*, *Parapharyngodon senisfaciecaudus cubensis*, *Aplectana hylambatis*, *Cosmocerca parva*, *C. podicipinus*, *Cosmocercella phyllomedusae*, *Schrankiana formosula*, *Falcaustra mascula*). This study presents new host and geographic records and expands knowledge of helminth diversity in a critically threatened group of vertebrates.

Key words: Acanthocephala, Nematoda, Anura, Argentina.

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Introduction

Research on parasitic helminths affecting amphibians in Argentina has grown over the last two decades. Although the Argentine Chaco has been extensively studied regarding parasitic helminths of amphibians, there are still information gaps regarding unexamined host species (González et al., 2021). Collections have not yet been conducted in some large geographic areas, and many taxonomic identities remain to be elucidated, mainly about the larval stages found in these vertebrates.

A recent review of publications on amphibians in Argentina between 2010 and 2020 revealed biases regarding the geographic distribution of the studies, the topics addressed, and the most studied species (Pereyra et al., 2023). The authors concluded that parasitological surveys were carried out mainly in the provinces of Corrientes and Buenos Aires, while records of helminths in amphibians in the remaining provinces are scarce. It is worth noting that Morphology, Ecotoxicology, and Diversity and Distribution were the thematic areas with the highest number of studies conducted. In addition, the most studied amphibian species were *Rhinella arenarum* (Hensel, 1867), *Boana pulchella* (Duméril & Bibron, 1841), *Leptodactylus latinasus* Jiménez de la Espada, 1875, *L. luctator* (Hudson, 1892), and *L. macrosternum* Miranda-Ribeiro, 1926. These results represent a starting point to elucidate and highlight the various biases in the scientific knowledge of Argentine anurans. Thus, parasitology remains an understudied research topic in our country compared to the rest of the thematic areas.

This study aims to identify the helminth fauna of twelve anurans species from Argentina's Dry and Humid Chaco ecoregions. Thus, we also provide new host and geographic records for eleven species of anurans from the Argentine Chaco, a threatened region whose hidden biodiversity, including helminth parasites, is still poorly known and studied.

Material and Methods

A total of 152 adult amphibians of 12 species belonging three families and five subfamilies were collected in three sites from two ecoregions in Argentina, between February 1997 and April 2018: Site a: approximately at 15 km northeastern of Corrientes city, Capital department, Corrientes province (27°42'87" S; 58°75'85" W), from now on referred to as "Corrientes", in the Humid Chaco ecoregion; Site b: approximately at 15 km eastern of Taco Pozo city, Almirante Brown department, Chaco province (25°35'21.7" S; 63°08'8.7" W), after this referred to as "Taco Pozo", and Site c: approximately 32 km south of Ingeniero Juárez city, Matacos department, Formosa province (24°21'60" S; 61°97'94" W) after this referred to as "Ingeniero Juárez" in the Dry Chaco ecoregion (Table 1; Fig. 1).

Amphibians were hand-captured between 18 and 21 hours using the sampling technique defined as "visual encounters survey" (Crump & Scott Jr., 1994) and "audio strip transect" method (Zimmerman, 1994) in the most favorable environments such as shallow ponds, temporary puddles, and ditches. Anurans were subsequently transported alive to a field-mounted laboratory, euthanised topically using 10% lidocaine cream, and necropsied. Under a stereoscopic microscope, we examined the

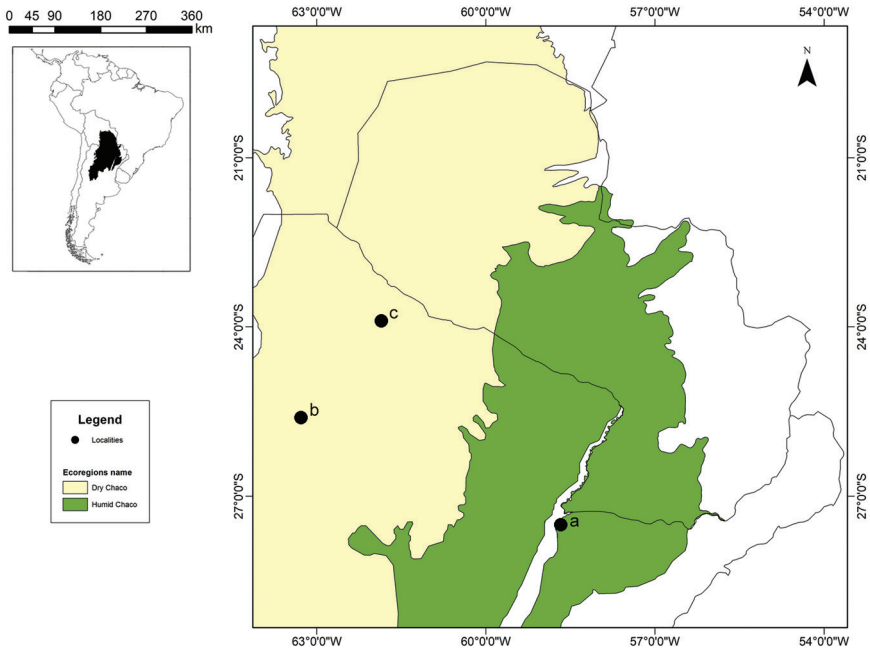


Fig. 1. Geographic location of the study sites in Humid and Dry Chaco ecoregions, Argentina; a — Corrientes, Corrientes province; b — Taco Pozo, Chaco province; c — Ingeniero Juárez, Formosa province

Table 1. Sample size (n) and collection site for twelve amphibian species from Dry and Humid Chaco, Argentina

Host species	Site a. n	Site b. n	Site c. n
Leptodactylidae: Leiuperinae			
<i>Physalaemus albonotatus</i> (Steindachner, 1864)			11
<i>Physalaemus santafecinus</i> Barrio, 1965	14		
Leptodactylidae: Leptodactylinae			
<i>Leptodactylus bufonius</i> Boulenger, 1894		3	
<i>Leptodactylus latinasus</i> Jiménez de la Espada, 1875			16
<i>Leptodactylus luctator</i> (Hudson, 1892)	14		
<i>Leptodactylus macrosternum</i> Miranda-Ribeiro, 1926	12		
Microhylidae: Gastrophryninae			
<i>Elachistocleis haroi</i> Pereyra, Akmentins, Laufer & Vaira, 2013		9	17
Hylidae: Hylinae			
<i>Dendropsophus nanus</i> (Boulenger, 1889)	17		
<i>Scinax nasicus</i> (Cope, 1862)	15		
<i>Pseudis platensis</i> (Gallardo, 1961)	10		
<i>Trachycephalus typhonius</i> (Linnaeus, 1758)	3		4
Hylidae: Phyllomedusinae			
<i>Pithecopus azureus</i> (Cope, 1862)	7		

Note: a — Corrientes, Capital department, Corrientes province (Humid Chaco); b — Taco Pozo, Almirante Brown department, Chaco province (Dry Chaco); c — Ingeniero Juárez, Matacos department, Formosa province (Dry Chaco).

digestive tract, lungs, liver, gallbladder, kidneys, urinary bladder, coelomic cavity, and musculature. Parasites were observed *in vivo*, counted, and then fixed in hot 70% ethyl alcohol.

Nematodes were clarified in Amman's lactophenol, mounted on temporary slides, and examined under a light microscope; acanthocephalans stained with hydrochloric carmine, cleared in eugenol, and mounted permanently in Canada balsam. We used a Leica DM2500 microscope equipped with a drawing tube for morphological examination and measurements. For scanning electron microscopy, the helminths were dehydrated in ethanol series, critical-point dried, coated with gold-palladium, and examined using a JEOL JSM-5800LV (Jeol, Tokyo, Japan). Nematode taxonomy followed Anderson et al. (2009) and Gibbons (2010), and Acanthocephala taxonomy followed Nielsen (2012) and Amin (2013). Amphibian taxonomy followed Frost (2024). Specimens were preserved in 70% ethyl alcohol and deposited in the Helminthological Collection of Centro de Ecología Aplicada del Litoral (CECOAL), Corrientes, Argentina.

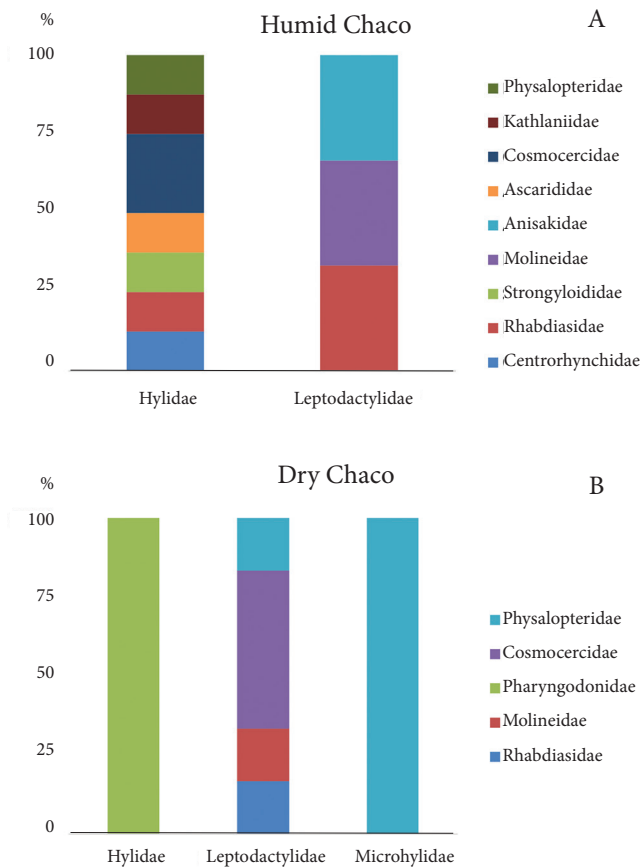


Fig. 2. Composition of helminth family in percentage, in each amphibian family analyzed from A — Humid Chaco and B — Dry Chaco, Argentina. Larva of Cystidicolidae family found in *P. albonotatus* from Dry Chaco was not incorporated in the figure; this record is considered an accidental infection

Results

We identified 17 helminth taxa parasitizing the lungs, small intestine, large intestine, and stomach serosa. These taxa were attributed to the class Acanthocephala and phylum Nematoda, distributed in 11 families. We identified ten helminth taxa from nine families in the Humid Chaco ecoregion, and eight taxa from five families in the Dry Chaco ecoregion. In the Humid Chaco, the hylid hosts showed the highest number of recorded helminth families, while in the Dry Chaco, leptodactylid hosts had the highest number of helminth families. Microhylids were collected only from the Dry Chaco, and we recorded a single family of helminths (Physalopteridae); similarly, the only hylid collected from the Dry Chaco, *T. typhonius*, was parasitised only by one species of pharyngodonid (Fig. 2, A, B).

Cosmocerids showed the highest species richness with four species, followed by Rhabdiasidae and Molineidae, each with two identified taxa. Larvae of genera *Centrorhynchus* Van Cleave, 1916 and *Physaloptera* Rudolphi, 1819 parasitized the most amphibian species. Cystacanths of *Centrorhynchus* were found in *D. nanus*, *P. azureus*, and *P. santafecinus*, and *Physaloptera* sp. in *P. azureus*, *L. luctator*, and *E. haroi*. On the other hand, *Physaloptera* sp. larvae were found in both ecoregions. *Leptodactylus luctator* was the amphibian species parasitized by the most significant number of helminth taxa. Table 2 shows the records of helminths found in this study by host and sampling site.

Acanthocephala

Order Polymorphida

Family Centrorhynchidae

Genus *Centrorhynchus* Van Cleave, 1916

Centrorhynchus sp. (cystacanth) (Fig. 3, A–C)

Site of infection: serous of stomach.

Host, locality, and collection date: *D. nanus*, Corrientes, Sep. 2004; *P. azureus*, Corrientes, Oct. 2008; *P. santafecinus*, Corrientes, Apr. 2003.

Voucher material: CECOAL 04090603 (*D. nanus*: 1 larva); CECOAL 08103002 (*P. azureus*: 1 larva); CECOAL 03043834 (*P. santafecinus*: 1 larva).

Comments: The proboscis of the cystacanths found in this study showed typical characteristics of the genus *Centrorhynchus*, including division into three regions, with the middle one inflated, and a constriction at the insertion of the proboscis receptacle. In addition, species of this genus were identified by the number of longitudinal rows, number of hooks per row, size of blades, and morphology and size of roots (Petrochenko, 1971).

Amphibian hosts can harbor both adult and larval stages of acanthocephalans. In the case of adults, males and females are found in the small intestines where they reproduce, with amphibians acting as definitive hosts (Arredondo & Gil de Pertierra, 2009). As for the larval stage, infective larvae can parasitize the celomic cavity attached to the serosa of various organs, such as the stomach, liver, intestinal wall, or muscles and mesenteries; and, in these cases, amphibians serve as paratenic hosts (Hamann et al., 2014).

In South America, the larval stage of this genus has been found in amphibians of the families Alsodidae, Bufonidae, Dendrobatidae, Hylidae, Leptodactyl-

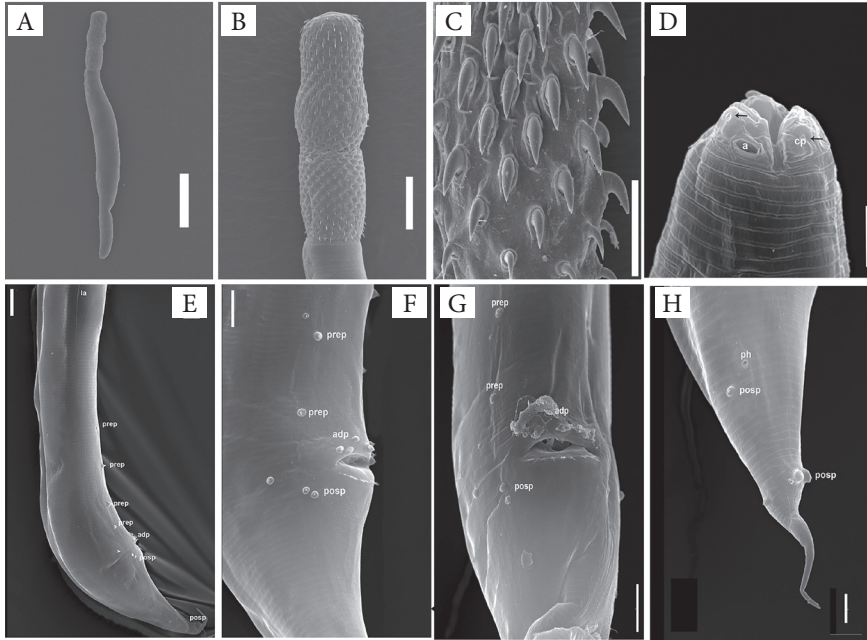


Fig. 3. Scanning electron micrographs of helminth parasites of Argentine amphibians. A–C — *Centrorhynchus* sp., cystacanth: A — whole worm; B — proboscis, lateral view; C — detail of hooks; D–H — *Schrankiana formosula*, male: D — anterior extremity, lateral view (black arrows, cp cephalic papillae); E — posterior extremity, lateral view (prep: precloacal papillae, adp: adcloacal papillae; posp: postcloacal papillae); F — detail of cloaca and precloacal, postcloacal, and adcloacal papillae, lateral view (prep: precloacal papillae, adp: adcloacal papillae; posp: postcloacal papillae); G — posterior extremity, detail of cloaca and precloacal, postcloacal and adcloacal papillae, ventral view (prep: precloacal papillae, adp: adcloacal papillae; posp: postcloacal papillae); H — posterior extremity, detail of postcloacal papillae and phasmid, lateral view (posp: postcloacal papillae, ph: phasmid). Scale bars: A: 250 μ m; B: 200 μ m; C: 100 μ m; D, H: 5 μ m; E: 20 μ m; F, G: 10 μ m

idae, Odontophrynidae, and Telmatobiidae (Campião et al., 2014; Santos et al., 2024). In Argentine amphibians, *Centrorhynchus* is a taxon with a broad host range; it has been found in *Melanophryniscus klappenbachii* Prigioni & Langone, 2000 (Bufonidae) from Chaco province, *Rhinella dorbignyi* (Duméril & Bibron, 1841) (= *R. fernandezae*), *Leptodactylus bufonius*, *L. macrosternum* (= *L. chaquensis*), *L. latinasus*, *Physalaemus cristinae* Cardozo, Tomatis, Dupont-Bru, Kolenc, Borteiro, Pansonato, Confalonieri, Lourenço, Haddad & Baldo, 2023 (= *P. albonotatus*) (Leptodactylidae), and *Scinax nasicus* (Hylidae) from Corrientes province (Hamann & González, 2015; González et al., 2019). Hamann & Kehr (1998) also found larval stages of acanthocephalans, registered as *Acanthocephala* gen. sp. in the hylid *D. nanus* (= *Hyla nana*) in Corrientes province. This study is the first record of *Centrorhynchus* sp. parasitizing *D. nanus*, *P. azureus*, and *P. santafecinus* from Argentina.

Nematoda**Class Secernentea****Order Rhabditida****Family Rhabdiasidae****Genus *Rhabdias* Stiles & Hassall, 1905*****Rhabdias* sp.**

Site of infection: lungs.

Host, locality, and collection date: *T. typhonius*, Corrientes, Oct. 2008.

Voucher material: CECOAL 08102202 (5 hermaphrodite specimens).

Comments: The arrangement and number of circumoral structures are among the main morphological characteristics used to differentiate *Rhabdias* species. In the Neotropical region, three patterns of oral arrangement are observed: (1) four submedian lips and two lateral pseudo lips; (2) six equal lips; and (3) absence of lips (Müller et al., 2018; Willkens et al., 2020). The specimens found in this study presented morphological and morphometric characteristics similar to *R. pseudosphaerocephala* that belongs to the first group, e. g., each submedian lip and pseudolabium with small papilla and amphids situated on pseudolabia, buccal capsule funnel-shaped, and esophagus club-shaped with dilation on anterior muscular part. The measurements of specimens studied here were slightly different from those reported by Kuzmin et al. (2007) for this species, e. g. total body length 6.22–9.10 mm (vs. 6.17–9.6 mm); body width 310–400 µm (vs. 290–380 µm), esophagus length 370–475 µm (vs. 400–460 µm), and esophagus length as percentage of body length 5.2–6.9 (vs. 4.4–6.5) (see Kuzmin et al., 2007). In this context, Müller et al. (2018) stated that *Rhabdias* species from the Neotropical region present cryptic diversity, and two species complexes, *R. breviensis* and *R. pseudosphaerocephala*, were observed based on molecular, taxonomic, ecological, and geographic data. Thus, to confirm the specific identification of the present specimens, molecular analyses using genetic markers, internal transcribed spacer (ITS) region (ITS1 + 5.8S + ITS2), and cytochrome oxidase I (COI) should be performed.

The life cycle of rhabdiasids presents two generations: a monoecious parasitic generation, with hermaphrodite adult females, and a free-living generation, typically dioecious, which includes adult males and females. This cycle can be either homogonic or heterogonic. Rhabdiasid nematodes that parasitize amphibians undergo homogonic cycles (Tkach et al., 2014).

In South America, the only reports of the genus *Rhabdias* in *T. typhonius* correspond to findings observed in the Atlantic Forest (da Graça et al., 2017) and in the Argentine Chaco, in the province of Formosa (Draghi et al., 2015a). Thus, we present the second report of *Rhabdias* in this amphibian host in Argentina and the first from Corrientes province.

***Rhabdias elegans* Gutiérrez, 1945**

Site of infection: lungs.

Host, locality, and collection date: *L. bufonius*, Taco Pozo, Oct. 2011.

Voucher material: CECOAL 11101910 (2 hermaphrodite specimens).

Comments: The specimens found here resemble *Rhabdias elegans* described in *Rhinella arenarum* (Hensel, 1867) (= *Bufo arenarum*) from Buenos

Aires and Tucumán provinces (Gutiérrez, 1945; Ramallo et al., 2020), and in *L. bufonius* from Corrientes province, Argentina (González & Hamann, 2008). This species belongs to the group of rhabdiasids that lack labial structures (Müller et al., 2018; Willkens et al., 2020). We also observed other morphological and morphometric characteristics to assign the specimens found here to *Rhabdias elegans*, which resemble specimens collected in Argentina, e. g., total body length (4.49–4.9 mm in specimens here study; 6.2 mm, 4.55–9.5 mm, 5.0–7.0 mm in specimens collected from Corrientes, Buenos Aires, and Tucumán provinces, respectively); length of the esophagus (350–390 µm in specimens here study; 390 µm, 314–490 µm, 310–400 µm in specimens collected from Corrientes, Buenos Aires, and Tucumán provinces, respectively); slightly post-equatorial position of the vulva (2.2–2.9 mm in specimens here study; 2.7 mm, 2.4–4.4 mm, 2.4–3.2 mm from posterior extremity in specimens collected from Corrientes, Buenos Aires, and Tucumán provinces, respectively) (Gutiérrez, 1945; González & Hamann, 2008; Ramallo et al., 2020).

In South America, *R. elegans* has been found in amphibians of families Bufonidae, Hylidae, Leptodactylidae, and Odontophrynidae (Campião et al., 2014). In Argentina, specifically, this species has been found in *R. arenarum* from Buenos Aires, Salta, and Tucumán provinces (Gutiérrez, 1945; Ramírez et al., 1979; Ramallo et al., 2020), in *L. bufonius* from Corrientes province (Hamann et al., 2012), in *R. major* from Chaco province (Hamann & González, 2015), and in *L. latrans* and *R. dorbignyi* (= *R. fernandezae*) from Buenos Aires province (Draghi et al., 2020 a). Our study is the first report of *R. elegans* in *L. bufonius* from Chaco province.

Family Strongyloididae

Genus *Strongyloides* Grassi, 1879

Strongyloides pereirai Travassos, 1932 (Fig. 4, A–C).

Site of infection: small intestine.

Host, locality, and collection date: *S. nasicus*, Corrientes, Oct. 2002.

Voucher material: CECOAL 02103042 (1 female).

Comments: This species was described by Travassos (1932) from specimens collected from *Hylodes nasicus* (Lichtenstein, 1823) (= *Elosia nasicus*) in Rio de Janeiro, Brazil. We used metric and morphological characteristics such as total length, esophagus length, distance of vulva from posterior end (3:1 ratio), length and width of eggs, and rounded posterior end to identify the specimens in this study.

In the life cycle of these nematodes, free-living males and females develop into adults in the soil, where they reproduce. Females lay eggs that hatch and develop into the L3 stage, which infects the vertebrate host. Following a migration through the host's body, adult parasitic females are found in the intestine (Viney & Lok, 2015).

In South America, nematodes of this genus have been found in amphibians from the families Bufonidae, Hylodidae, and Leptodactylidae (Campião et al., 2014). In Argentina, they were previously found in *P. cristinae* (= *P. albonotatus*) from Corrientes province (González & Hamann, 2012a). This is the first report of *S. pereirai* from Argentine amphibians.

Order Strongylida**Family Molineidae****Genus *Oswaldocruzia*** Travassos, 1917***Oswaldocruzia proencai*** Ben Slimane & Durette-Desset, 1995

Site of infection: small intestine.

Host, locality, and collection date: *L. luctator*, Corrientes, Feb. 1997.

Voucher material: CECOAL 97020542 (1 male).

Comments: This nematode genus parasitizes the intestines of amphibians and reptiles worldwide (Willkens et al., 2021). Lent et al. (1946) reported *O. mazzai* in *Rhinella diptycha* (= *Bufo paracnemis*), *Leptodactylus luctator* (= *L. ocellatus*), and *L. bufonius* from Asunción, Chaco-i, and Remanso Castillo, Paraguay. However, it was later suggested that these specimens be reassigned to *O. proencai* (Ben Slimane & Durette-Desset, 1995). This species is characterized by having a divided cephalic vesicle; in males, the caudal bursa is Type II, with ray 8 arising above the root of the dorsal ray and overlapping ray 6 for only half its total length. In addition, the dorsal ray is conical and shares its base with ray 8. Additionally, the spicular fork dividing more distally, with both branches of the fork of equivalent length; the blade divides distally into 10 branches (Ben Slimane & Durette-Desset, 1995).

The life cycle of nematodes of this genus is monoxenous (Anderson, 2000). Baker (1978) and Hendrikx (1983) studied the life cycles of *O. filiformis* (Goeze, 1782) Travassos, 1917 and *O. pipiens* Walton, 1929, respectively, and established that infection occurs orally through ingestion of infective larvae or through penetration of infective larvae upon contact with the skin of the amphibian.

In South America, *O. proencai* has been found in amphibians of families Bufonidae, Hylidae, Leptodactylidae, and Strabomantidae (Campião et al., 2014). In Argentina, specifically, *O. proencai* has been found in *R. diptycha* (= *R. schneideri*) from Corrientes province (González & Hamann, 2008) and in *R. arenarum* and *R. diptycha* from Salta province (Ramallo et al., 2007). This is the first report of *O. proencai* in *L. luctator* from Argentina.

Oswaldocruzia subauricularis (Rudolphi, 1819) Travassos, 1917 (Fig. 4, D–G)

Site of infection: small intestine.

Host, locality, and collection date: *P. albonotatus*, Ingeniero Juárez, Feb. 2018; *L. latinasus*, Ingeniero Juárez, Apr. 2018.

Voucher material: CECOAL 18021903 (*P. albonotatus*: 2 females, 1 male); CECOAL 18042439 (*L. latinasus*: 1 female, 2 males).

Comments: This species is characterized by a divided cephalic vesicle; a synlophe with cuticular ridges supported by chitin throughout the entire body; vulva without protruding lips; the bursal rays 6, 8, and dorsal ray Type III; spicules formed from three branches: “fork”, “blade”, and ‘shoe’; fork divided below its posterior third into two branches of equivalent length but different shapes (Ben Slimane & Durette-Desset, 1995). We also observed morphometric characteristics which resemble specimens collected by Ben Slimane & Durette-Desset (1995) as total body length (8.5–9.1 mm vs. 8.2–9.6 mm); width in the middle part of the body (170–175 µm vs. 160–180 µm); cephalic vesicle length (92–100 µm vs. 85–95 µm); cephalic vesicle wide (78–85 µm vs. 85–95 µm in its swollen part; 46–48 µm vs. 40–45 µm in its thin part); nerve ring, excretory pore and

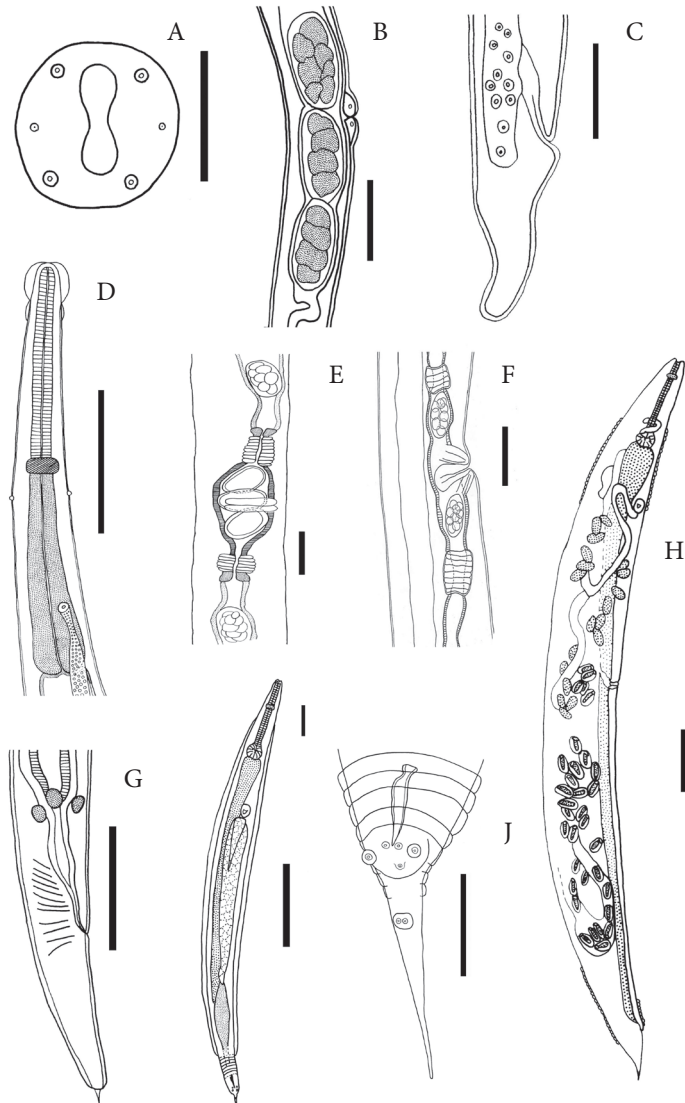


Fig. 4. Helminth parasites of Argentine amphibians. A–C — *Strongyloides pereirai*, female: A — anterior extremity, apical view; B — detail of vulva and eggs, lateral view; C — posterior extremity, lateral view; D–G — *Oswaldocruzia subauricularis*, female: D — anterior extremity, ventral view; E — detail of reproductive system, ventral view; F — detail of reproductive system, lateral view; G — posterior extremity, lateral view; H–J — *Parapharyngodon senisfaciecaudus cubensis*: H — female, whole worm, lateral view; I — male, whole worm, ventral view; J — male, detail of posterior extremity, ventral view. Scale bars: A: 10 μ m; B, J: 50 μ m; C: 30 μ m; D: 150 μ m; E, F, G: 100 μ m; H, I: 500 μ m

deirids to the anterior end (210–230 μ m, 420–450 μ m, 487–500 μ m vs. 185–230 μ m, 380–445 μ m, and 420–495 μ m, respectively); esophagus length (450–520 μ m vs. 480–560 μ m); spicules length (175–205 μ m vs. 190–210 μ m) in males; total body length (14.0–17.6 mm vs. 14.45 mm–18.0 mm); width in the middle part of body (220–240 μ m vs. 190–230 μ m); cephalic vesicle length (100–120 μ m vs. 95–130 μ m); cephalic vesicle wide (50–55 μ m vs. 45–60 μ m in its swollen part; 42–52 μ m vs. 40–50 μ m in its thin

part); nerve ring, excretory pore and deirids to the anterior end (230–250 μm , 380–450 μm , 450–520 μm vs. 245–280 μm , 390–530 μm , and 420–580 μm , respectively); esophagus length (520–590 μm vs. 500–640 μm); vulva and anus to posterior end (5.2–6.1 mm and 190–205 μm vs. 4.45–6.5 mm and 185–210 μm , respectively); size of eggs (90–95 μm x 50–52 μm vs. 85–100 μm x 50–60 μm) in females.

In South America, *O. subauricularis* has been found in amphibians from the families Bufonidae, Ceratophryidae, Hylidae, and Leptodactylidae (Campião et al., 2014). In Argentina, specifically, this species has been found in *R. diptycha* from Formosa province, and in *R. dorbignyi* (= *R. fernandezae*) from Corrientes province (González et al., 2021; Hamann et al., 2013 a). This is the first report of *O. subauricularis* in *P. albonotatus* and in *L. latinasus*.

Order Oxyurida

Family Pharyngodonidae

Genus *Parapharyngodon* Chatterji, 1933

Parapharyngodon senisfaciecaudus cubensis Barus & Coy Otero, 1969 (Fig. 4, H–J).

Site of infection: large intestine.

Host, locality, and collection date: *T. typhonius*, Ingeniero Juárez, Feb. 2017.

Voucher material: CECOAL 17022105 (2 females, 1 male).

Comments: These specimens were identified based on the length of males and females, the ovary surrounding the esophagus, the length of the spicule, the number and arrangement of caudal papillae, the absence of an echinate cloacal lip, and the extension of lateral alae in males. The caudal papillae of the males have the same number and arrangement as those described by Barus & Coy Otero (1969) for lizards of the family Anolidae: one pair of precloacal papillae, one pair of adcloacal papillae, one pair of papillae in the first third of the caudal filament, and one unpaired postcloacal papilla.

Members of the order Oxyurida are strictly monoxenous (Anderson, 2000); eggs released into the environment can be a source of oral infection when they contaminate the prey consumed by amphibians.

Parapharyngodon spp. previously found in this amphibian host include *P. duniae* Bursey & Brooks, 2004 from Costa Rica, *P. hylidae* Velarde-Aguilar, Mata-López, Guillén-Hernández & León-Règagnon, 2015 from México, and *P. hugoi* Pereira, Campião, Luque & Tavares, 2017 from Brazil (Bursey & Brooks 2004; Velarde-Aguilar et al., 2015; Pereira et al., 2017). This is the first report of *P. senisfaciecaudus cubensis* from Argentinean amphibians.

Order Ascaridida

Family Anisakidae

Genus *Contracaecum* Ralliet & Henry, 1912

Contracaecum sp. (larva) (Fig. 5, A–C)

Site of infection: gastric mucosa.

Host, locality, and collection date: *L. luctator*, Corrientes, Mar. 2002.

Voucher material: CECOAL 02032605 (1 larva)

Comments: This larva was identified at the genus level based on the following features: the esophagus with a ventriculus, the presence of a posterior ventricular

appendix and an anterior intestinal caecum, the location of the excretory pore at the base of the lip, and the presence of a tooth-like structure on the ventrolateral lip. According to Anderson et al. (2009), these are the main morphological traits used to identify nematodes of this genus.

Contracaecum spp. has definitive hosts, including various species of piscivorous birds and mammals of freshwater, brackish, and marine environments. Their eggs are excreted in feces and enter water bodies, where they undergo embryonic development into the L1 stage. Then, they move to L2 after molting. These eggs or larvae can be ingested by first intermediate hosts and grow within their hemocoel. Different species of invertebrates can serve as primary, and intermediate hosts; their precise roles in facilitating larval transmission to fish intermediate hosts remain partially understood. Upon ingesting infected invertebrates by second intermediate hosts, the larvae molt to the L3 stage. Various teleost fishes can act as second intermediate or paratenic hosts. Definitive hosts become infected by preying upon the second intermediate or paratenic hosts (Shamsi, 2019). Additionally, amphibians can act as paratenic hosts in the life cycle of this nematode, although the occurrence of this genus in amphibians is sporadic.

Third-stage larvae have been found in *Siren nettingi* Goin, 1942 (= *Siren intermedia texana*) from USA (McAllister & McDaniel, 1992), in *Osteopilus septentrionalis* (Duméril & Bibron, 1841) and in *Aquarana catesbeiana* (Shaw, 1802) (= *Rana catesbeiana*) from Cuba (Coy Otero & Ventosa, 1984), and in *Xenopus laevis* (Daudin, 1802) from Chile (Castillo et al., 2017). In Argentina, *Contracaecum* sp. has been found in *D. nanus* (= *Hyla nana*), from Corrientes province (Hamann & Kehr, 1998), and in introduced species *A. catesbeiana* (= *L. catesbeianus*) from San Juan province (González et al., 2014). This is the first report of *Contracaecum* sp. in *L. luctator* from Argentina.

Family Ascarididae

Genus *Porrocaecum* Ralliet & Henry, 1912

Porrocaecum sp. (larva) (Fig. 5, D, E)

Site of infection: liver.

Host, locality, and collection date: *T. typhoni*, Corrientes, Oct. 2008.

Voucher material: CECOAL 08102201 (1 larva)

Comments: The larvae of this genus have an esophagus with an anterior muscular portion and a posterior portion known as the ventriculus, which is oblong in the specimen studied here. They do not have an esophageal appendix. In addition, they have an intestinal caecum that is as broad as or broader than the esophagus, with variable length, which extends towards the anterior extremity and is located dorsally to the esophagus (Anderson et al., 2009). Given that the specimen analyzed in this study presented the characteristics mentioned above, we consider that it belongs to the genus *Porrocaecum*.

In the life cycle of *Porrocaecum* spp., the eggs are released into the environment through the feces of land birds, the definitive hosts. The eggs are then ingested by annelids, the intermediate host, where they develop into L3. Small mammals can consume these intermediate hosts, thus becoming paratenic hosts and transmitting the parasite to carnivorous hosts. Some species of ducks, passerines, and other small birds do not normally consume these small mammals, so the paratenic

vertebrate host is eliminated from the transmission cycle. In such cases, the definitive host becomes infected directly by ingesting the invertebrate (Anderson, 2000). Bursey et al. (2001) suggested that amphibians can fulfill the role of transport hosts in this cycle.

In South America, larvae of the genus *Porrocaecum* have been found in amphibians of families Bufonidae, Dendrobatidae, Hylidae, Leptodactylidae, and Pipidae (Campião et al., 2014, 2016). In Argentina, this taxon has been previously found in *R. diptycha* and *L. macrosternum* from Corrientes province, and in *R. major* from Formosa province (González et al., 2021). This is the first report of the genus *Porrocaecum* in *T. typhoni*us.

Family Cosmocercidae

Genus *Aplectana* Railliet & Henry, 1916

Aplectana hylambatis (Baylis, 1927) Travassos, 1931

Site of infection: large intestine

Host, locality, and collection date: *L. latinasus*, Ingeniero Juárez, Apr. 2018.

Voucher material: CECOAL 18042430 (10 females, 10 males).

Comments: This species was described from specimens collected from *Leptopelis aubry* (Duméril, 1856) (Arthroleptidae) in Guinea, Africa (Baylis, 1927). The main morphological characteristic is the possession of spicules with articulation in their distal part, giving them the appearance of a 'hockey stick.' Additionally, females exhibit modifications of the cuticle around the vulva, characterized by mamelon-like protuberances that vary in number, size, and arrangement (Baker, 1980). The specimens examined in this study resemble those found in *L. bufonius* from Taco Pozo, as well as *R. arenarum* from both La Plata and San Juan, Argentina, particularly in the number and arrangement of male caudal papillae (4 : 5 : 6 +1). However, these specimens differ from those collected in *R. arenarum* from La Plata, which exhibit two rows of somatic papillae between the precloacal papillae pairs — an arrangement not observed in the specimens studied here. Additionally, they differ from those found in the same host from San Juan in the positioning of the 4th, 5th, and 6th pairs of postcloacal papillae (ventral, lateral, and ventral, respectively). The arrangement of the postcloacal papillae in the specimens studied here corresponds with that found in *L. bufonius* from Taco Pozo: the 1st small pair is ventral, the 2nd and 3rd pairs are larger and ventrolateral and are located at the middle of the tail approximately, the 4th and 5th pairs are lateral, and the 6th pair is ventral (González et al., 2019). Additionally, the lengths of the spicules and gubernaculum (230–250 µm and 52–60 µm vs. 170–246 µm and 50–73 µm, respectively) and the presence of the single small, medial mamelon-like cuticular protuberance on the anterior lip of vulva also resemble the specimens collected in *L. bufonius* from Taco Pozo, Argentina (González et al., 2019).

The cosmocercids of this genus have a monoxenous life cycle; amphibians become infected by ingesting eggs with the infective stage (Anderson, 2000).

Aplectana hylambatis is a species with a wide geographic and host distribution worldwide (Baker, 1980); in South America, it has been found in different amphibian

families in seven countries (Campião et al., 2014). In the South American Chaco, *A. hylambatis* is the species with the most extensive geographic range and host records (González et al., 2021); specifically, in Argentina it has been found in eight provinces parasitizing anurans of the families Bufonidae, Ceratophryidae, Hylidae, Leptodactylidae, Microhylidae, and Odontophrynidae Hamann et al., 2022 (González et al., 2021; Hamann et al., 2022; Piñeiro Gómez et al., 2023). *Aplectana hylambatis* has been previously found in *L. latinasus* from Corrientes province (González & Hamann, 2015), so this is the first report on this leptodactylid from Formosa province. Considering the broad geographic distribution of *A. hylambatis*, further integrative taxonomic analyses would be necessary to determine whether this species is widely distributed in South America or represents a species complex.

Genus *Cosmocerca* Diesing, 1861

Cosmocerca parva Travassos, 1925 (Fig. 5, I, J)

Site of infection: large intestine.

Host, locality, and collection date: *L. luctator*, Corrientes, Sep. 1998.

Voucher material: CECOAL 98091369 (2 females, 2 males).

Comments: *Cosmocerca parva* was described from *Hylodes nasus* (Lichtenstein, 1823) (= *Elosia nasus*) in Rio de Janeiro, Brazil (Travassos, 1925). Like all species of the genus, it is characterized by the presence of rosette papillae supported by plectanes, the presence of two spicules, the absence of caudal alae in males, and the presence of two prodelphic ovaries in females (Gibbons, 2010). In this species, the number of papillae varies between host species and collection locations (González et al., 2023). Generally, males of this species present 4 to 7 pairs of precloacal papillae, 2 to 4 pairs of adcloacal papillae, and 3 pairs of postcloacal papillae. Regarding the adcloacal papillae, some specimens possess a single median papilla located on the anterior lip of the cloaca (Mordeglia & Digiani, 1998), while is absent in other specimens (González et al., 2023). The specimens examined in this study present the following arrangement of caudal papillae: six precloacal pairs, first four pairs with plectanes; plectanes consist of a central papilla surrounded by small punctations and supported by anterior and posterior sclerotized plates; fifth and sixth pairs with one inner and one outer circle of 11–12 punctations, without plectanes. Three pairs of adcloacal papillae, one anterior, one lateral, and one posterior to the cloaca, absence of small unpaired median papilla on the anterior lip of the cloaca. Three pairs of postcloacal papillae, first and third pairs lateral, and second pair ventral. Compared with specimens collected in the Argentine Chaco, the presence of 6 pairs of precloacal papillae corresponds to the specimens studied in *R. major* (= *Bufo granulatus major*), *P. santafecinus*, *S. acuminatus*, *P. cristinae* (= *P. albonotatus*), and *L. podicipinus*; while the absence of anterior papilla to the cloaca is similar to specimens collected in *Rhinella bergi* from Corrientes (Mordeglia & Digiani, 1998; González & Hamann, 2007; 2008; 2012 a; 2016).

Amphibians become infected with nematodes of this genus by active penetration of L3, which occurs in the environment through the conjunctiva of the hosts' eyes (Kirillova & Kirillov, 2021).

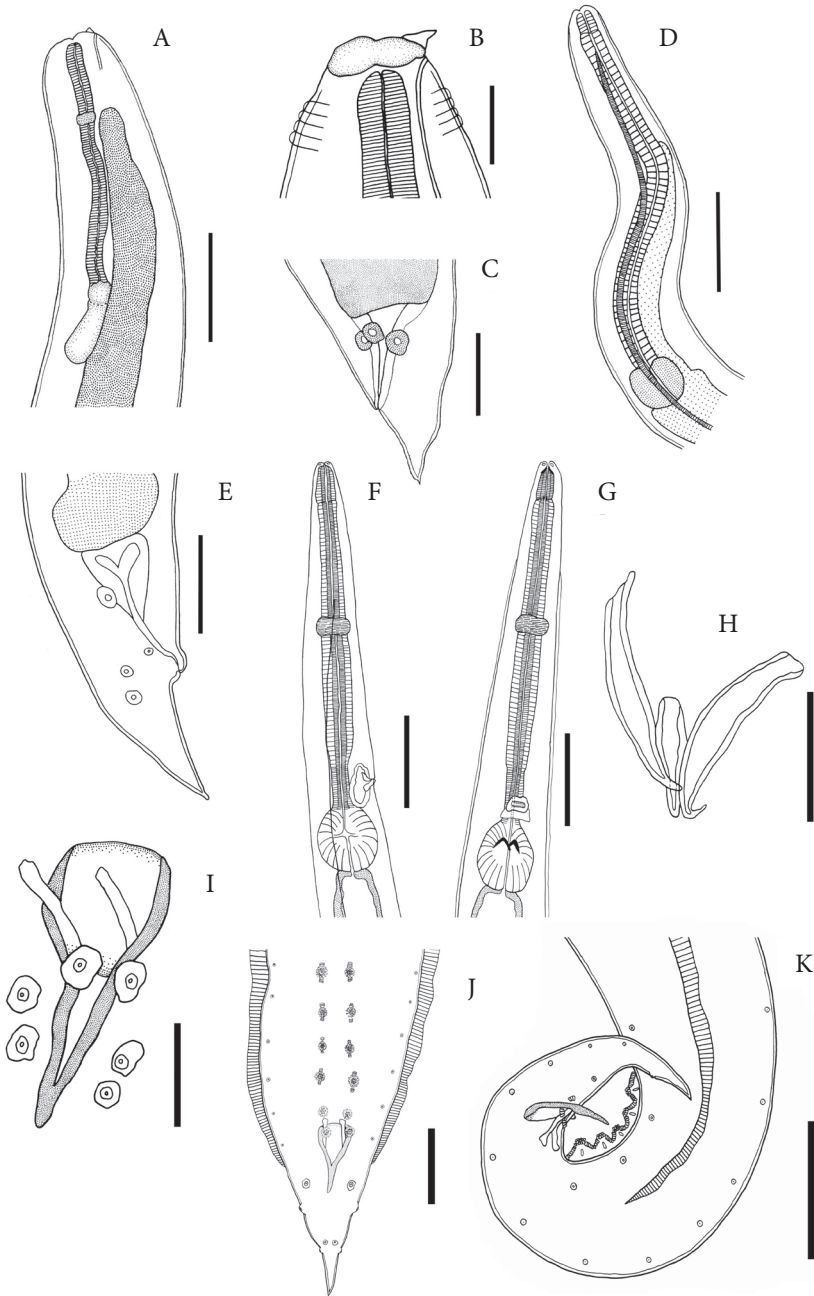


Fig. 5. Helminth parasites of Argentine amphibians. A–C — *Contracaecum* sp., larva: A — anterior extremity, lateral view; B — detail of anterior extremity, lateral view; C — posterior extremity, lateral view; D–E — *Porrocaecum* sp., larva; D — anterior extremity, lateral view; E — posterior extremity, lateral view; F–H — *Schrankiana formosula*: F — male, anterior extremity, lateral view; G — female, anterior extremity, ventral view; H — detail of spicules and gubernaculum; I–J — *Cosmocerca parva*, male: I — detail of spicules, gubernaculum and adcloacal papillae, ventral view; J — posterior extremity, ventral view; K — *Cosmocerca podicipinus*, male, posterior extremity, lateral view. Scale bars: A: 500 μm ; B: 40 μm ; C: 200 μm ; D: 300 μm ; E, F, G, J, K: 100 μm ; H: 40 μm ; I: 30 μm .

This species has a vast host and geographic range; it has been found in six countries in South America, infecting amphibians from families Bufonidae, Craugastoridae, Dendrobatidae, Hylidae, Hylodidae, Leptodactylidae, and Odontophrynidae (Campião et al., 2014). In Argentina, *C. parva* is the most common adult nematode in amphibian hosts (González & Hamann, 2015). Draghi et al. (2020 a) previously found *C. parva* in *L. luctator* (= *L. latrans*) from Buenos Aires province; this is the first report of this cosmocercid in *L. luctator* from Corrientes province. Considering the broad geographic distribution of *C. parva*, it would be necessary to perform further integrative taxonomy analyses to assess if this species is widely distributed in South America or represents a species complex.

Cosmocerca podicipinus Baker & Vaucher, 1984 (Fig. 5, K)

Site of infection: large intestine.

Host, locality, and collection date: *P. albonotatus*, Ingeniero Juárez, Feb. 2017–Apr. 2018.

Voucher material: CECOAL 17022148 (1 female); CECOAL 18042432 (1 male).

Comments: This species was described from specimens collected from Paraguayan leptodactylids by Baker & Vaucher (1984). We easily identified this species from other species of the genus because the precloacal papillae, with plectanes, show no variation in number (there are five pairs) and are fused by a chitinous support that joins the papillae in the same row.

In the South American Chaco, *C. podicipinus* was the nematode species that parasitized the most host species (20 species) (González et al., 2021). Previously, this cosmocercid has been found in *P. cristinae* (= *P. albonotatus*) from Corrientes province (González & Hamann, 2012a); this is the first record from *C. podicipinus* in *P. albonotatus* from Dry Chaco.

Genus *Cosmocercella* Steiner, 1924

Cosmocercella phyllomedusae Baker & Vaucher, 1983 (Fig. 6, A–E)

Site of infection: large intestine.

Host, locality, and collection date: *P. azureus*, Corrientes, Nov. 2008.

Voucher material: CECOAL 08112503 (5 females, 5 males).

Comments: This species was described with specimens collected from *P. azureus* (= *Phyllomedusa hypochondrialis*) in Paraguay (Baker & Vaucher, 1983). This is unique among the species of *Cosmocercella* due to the following characteristics precloacal papillae: four large unpaired vesiculated papillae, markedly large rosette precloacal papillae surrounded by broad areas of cuticular punctations, and small rosette papillae. In addition, it features three adcloacal papillae, one medial unpaired papilla, and a pair on both sides of the anterior cloacal lip. The tail has a pair of prominent subventral papillae surrounded by a raised rosette of bosses. Females lack somatic papillae (Baker & Vaucher, 1983).

The life cycle of nematodes belonging to this genus remains unknown. Probably, like other cosmocercids, it has a direct life cycle (Anderson, 2000). Further research is needed to determine whether infection occurs through penetration of the L3 stage or ingestion of eggs.

Bursey et al. (2001) reported this species from *Pithecopus palliatus* (Peters, 1873) (= *Phyllomedusa palliata*), *Callimedusa tomopterna* (Cope, 1868) (= *Phyllomedusa tomopterna*), and from *Phyllomedusa vaillantii* Boulenger, 1882 in Peru. Three species of this genus have been described up to the moment from Neotropical Realm, *C. anothecae* Baker & Adamson, 1977, *C. phyllomedusae* and *C. minor* (Freitas & Dobbin, 1961) Baker & Vaucher, 1983; of them, only the last one has been found in Argentine amphibians (González & Hamann, 2012 b). We provide the first record of *C. phyllomedusae* from Argentina.

Genus *Schrankiana* Strand, 1942

= *Schrankia* Travassos, 1925; = *Schrankinema* Travassos, 1949

Schrankiana formosula

= *S. inconspicata* Freitas, 1959 (Figs 3, D–H; 5, F–H)

Site of infection: large intestine.

Host, locality, and collection date: *L. latinasus*, Ingeniero Juárez, Dec. 2017.

Voucher material: CECOAL 17121116 (10 females, 10 males).

Comments: This species was described by Freitas (1959) from specimens collected from *Leptodactylus fuscus* (Schneider, 1799) (= *L. typhoni*) in Brazil. Recently, Félix et al. (2024) redescribed *S. formosula* based on re-examining type specimens and collecting material from *Leptodactylus pentadactylus* (Laurenti, 1768) in Amapá State, Brazil. Also, they re-examined the type series of *S. inconspicata* and proposed it as a junior synonym of *S. formosula*. Additionally, they established that *Schrankiana* is a member of the family Cosmocercidae, not Atractidae, based on morphological similarities and molecular phylogenetic analyses. The specimens analyzed here closely resemble those described by Baker & Vaucher (1988), exhibiting four pairs of subventral precloacal papillae; three pairs of adcloacal papillae in the anterior lip of the cloaca with one large unpaired papillae between them; and five pairs of postcloacal papillae: two pairs of adjacent subventral papillae in the anterior half of tail, and one pair lateral, one pair subventral, and one pair of subdorsal papillae in the posterior half of tail. In addition, the specimens had an elongated and slender pharynx, spicules less than 100µm long, and a gubernaculum less than 55µm long (Baker & Vaucher, 1988).

No information about the life cycle of this genus in amphibian hosts is currently available (Anderson, 2000).

Posteriorly its description, *S. formosula* has been found in Leptodactylidae from Paraguay, in Brachycephalidae, Leptodactylidae, and Hylidae from Brazil, in Leptodactylidae from Costa Rica, and Leptodactylidae from Peru (Baker & Vaucher, 1988; Souza Lins et al., 2017; Félix et al., 2024). In Argentina, *S. formosula* has been found in *L. bufonius* from Formosa province (González et al., 2021); so this is the second report of this species from Argentina and the first in *L. latinasus* from Ingeniero Juárez, Formosa province.

Family Kathlaniidae

Genus *Falcaustra* Lane, 1915

Falcaustra mascula (Rudolphi, 1819) Freitas & Lent, 1941 (Figs 6, F, G; 7, A–E)

Site of infection: large intestine.

Host, locality, and collection date: *P. platensis*, Corrientes, Nov. 2011; *L. lucator*, Corrientes, Dec. 1998–May. 2003; *L. macrosternum*, Corrientes, Mar.–Apr. 2003.

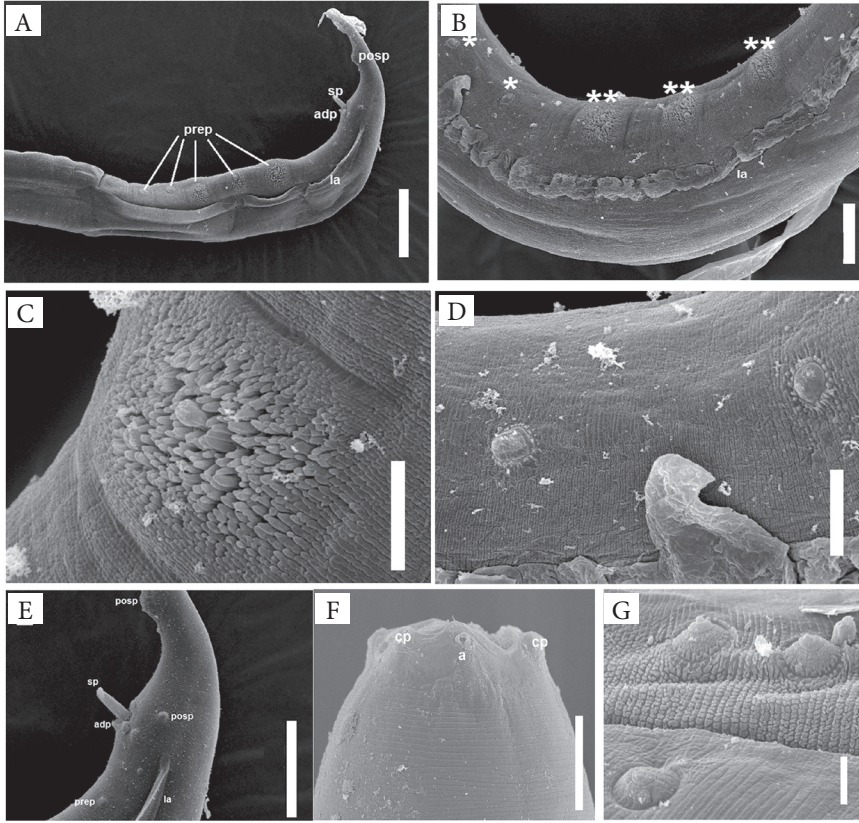


Fig. 6. Scanning electron micrographs of helminth parasites of Argentine amphibians. A–E — *Cosmocercella phyllomedusae*, male: A — posterior extremity, lateral view (prep: postcloacal papillae, adp: adcloacal papillae, posp: postcloacal papillae, sp: spicules, la: lateral alae); B — posterior extremity, detail of disposition of precloacal papillae, lateral view (one asterisk indicating simple precloacal papillae; two asterisk indicating precloacal papillae surrounded by cuticular punctations); C — detail of precloacal papillae surrounded by cuticular punctations; D — detail of simple precloacal papillae; E — detail of protruded spicule and adcloacal papillae, lateral view (prep: postcloacal papillae, adp: adcloacal papillae, posp: postcloacal papillae, sp: spicules, la: lateral alae); F–G — *Falcaustra mascula*, male: F — anterior extremity, lateral view (a: amphid, cp: cephalic papillae); G — detail of precloacal papillae, ventrolateral view. Scale bars: A: 100 μm ; B, F: 50 μm ; C: 25 μm ; D, E: 75 μm ; G: 10 μm

Voucher material: CECOAL 08112502 (*P. platensis*: 1 male); CECOAL 03040804 (*L. macrosternum*: 1 males); CECOAL 03054067 (*L. luctator*: 2 females).

Comments: This species is characterized by the presence of a pseudosucker, one pair of adcloacal papillae, six pairs of postcloacal papillae, the absence of an unpaired precloacal papilla, falcate spicules, and highly developed precloacal musculature in males. Additionally, the cuticle of nematodes in this genus displays transverse striations, and the mouth is surrounded by three large lips, each with bifurcated papillae. The esophagus exhibits a sub-spherical isthmus, and the excretory pore, located anteriorly to the isthmus, is very prominent (Gomes & Vicente, 1966).

Adults of *Falcaustra* spp. parasitize turtles, amphibians, fish, and one species found in birds (Baker, 1987). The life cycle of this species in amphibians is unknown. Third-stage larvae have been found in snails and fishes, suggesting that these organisms could act as paratenic host (Bartlett & Anderson, 1985).

In South America, *F. mascula* has been found in amphibians of families Brachycephalidae, Bufonidae, Hylidae, Hylodidae, Leptodactylidae, and Telmatobiidae (Campião et al., 2014; Chero et al., 2014; Toledo et al., 2015). In Argentina, two species of *Falcaustra* have been found up to the moment; *F. mascula* collected in *R. diptycha* (= *R. schneideri*) from Corrientes province, and *F. sanjuanensis* in *Odonophrynus* cf. *barrioi*, in *L. luctator* (= *L. latrans*), and in the introduced frog, *Aquarana catesbeiana* (= *L. catesbeianus*) from San Juan province (González et al., 2013; González & Hamann, 2015; Ramallo et al., 2016). The present work represents the first record of the genus *Falcaustra* in *P. platensis* and *L. macrosternum*.

Order Spirurida

Family Physalopteridae

Genus *Physaloptera* Rudolphi, 1819

Physaloptera sp. (larva) (Fig. 7, F, G)

Site of infection: serous of the stomach.

Host, locality, and collection date: *P. azureus*, Corrientes, Mar. 2008; *L. luctator*, Corrientes, Mar. 2003; *E. haroi*, Ingeniero Juárez, Dec. 2016; Taco Pozo, Oct. 2011.

Voucher material: CECOAL 08031801 (*P. azureus*: 1 larva); CECOAL 02032585 (*L. luctator*: 1 larva); CECOAL 16122123 (*E. haroi*: 2 larvae); CECOAL 11101923 (*E. haroi*: 1 larva).

Comments: The specimens studied here exhibit the same morphology as larvae of the same genus collected in previous studies as: transversely annulated cuticle; apical end with two lateral lips and a cephalic collar formed by inflated cuticle. Each lip, with a sclerotized support and one terminal tooth at the upper margin and with two cephalic papillae and one amphid. Glandular esophagus long, two and a half to three times the length of the muscular esophagus. Nerve ring surrounding the muscular esophagus in its posterior half; excretory pore at the level of the muscular esophagus (González & Hamann, 2007; 2008; 2012 a; Felix-Nascimento et al., 2022). These larvae are found attached to the gastric mucosa of amphibians by their cephalic collar.

Physaloptera sp. has a heteroxenous life cycle, parasitizing various vertebrates, with numerous invertebrates acting as intermediate hosts. These invertebrates ingest the eggs shed in the feces of definitive hosts. Inside the intestine of the intermediate host, a larva hatches and migrates to its tissues to continue developing into the L3 stage. These larvae infect definitive and paratenic hosts, such as amphibians (Anderson, 2000).

In South American amphibians, this genus is widely distributed and has been found in anurans of the families Aromobatidae, Bufonidae, Craugastoridae, Hylidae, Leptodactylidae, and Odontophrynidae (Campião et al., 2014; Toledo et al., 2015). In Argentine amphibians, this larva has been found in *Rhinella dorbignyi* (= *R. fernandezae*), *R. diptycha*, *Scinax acuminatus* (Cope, 1862), *S. nasicus*, *Physalaemus*

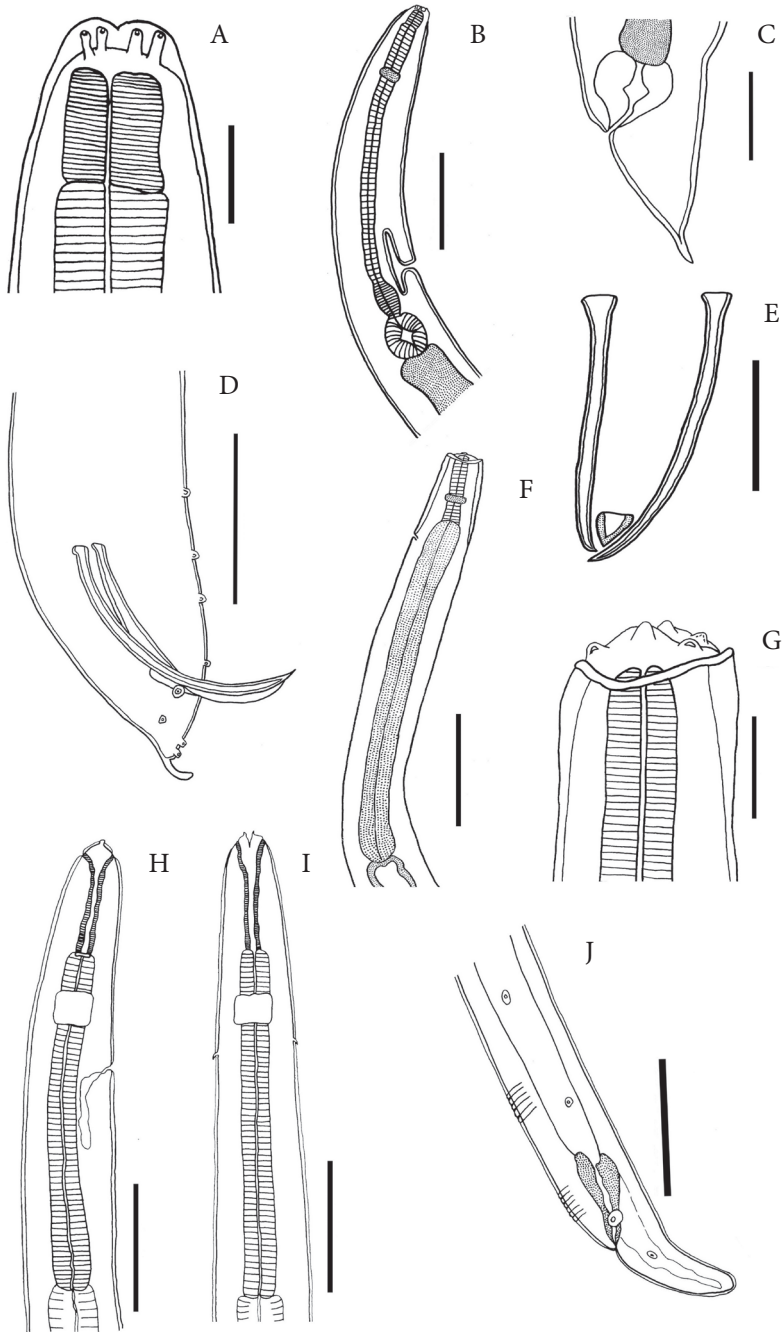


Fig. 7. Helminth parasites of Argentine amphibians. A–E — *Falcaustra mascula*: A — male, detail of anterior extremity, ventral view; B — female, anterior extremity, lateral view; C — female, posterior extremity, lateral view; D — male, posterior extremity, lateral view; E — detail of spicules and gubernaculum, ventral view; F–G: *Physaloptera* sp., larva; F — anterior extremity, lateral view; G — detail of pseudolabia and cephalic collar, ventral view; H–J. — larva aff. *Cystidicolidae*: H — anterior extremity, lateral view; I — anterior extremity, dorsal view; J — posterior extremity, lateral view. Scale bars: A, G: 50 μ m; B: 400 μ m; C, E: 200 μ m; D, F: 300 μ m; H, I, J: 100 μ m

santafecinus, *P. cristinae* (= *P. albonotatus*) from Corrientes province, in *R. major* from Corrientes, Chaco, and Formosa provinces, in *Leptodactylus bufonius* from Corrientes and Formosa provinces, in *Physalaemus biligonigerus* (Cope, 1861) from Córdoba province, in *Melanophryniscus klappenhabii* from Chaco province, and *Chacophrys pierottii* (Vellard, 1948) from Formosa province (Gutiérrez et al., 2005; González et al., 2021). We present here the first record of *Physaloptera* larvae in *P. azureus*, *L. luctator*, and *E. haroi* from Argentina.

Superfamily Habronematoidea

Family aff. Cystidicolidae (larva) (Fig. 7, H–J)

Site of infection: stomach wall.

Host, locality, and collection date: *P. albonotatus*, Ingeniero Juárez, Dec. 2016.

Voucher material: CECOAL 16122111 (1 larva).

Comments: The larva found in this study had a pseudolabia that did not cover the entire cephalic surface, an oral opening with well-defined bilateral symmetry, and an esophagus divided into anterior muscular and posterior glandular sections. Thus, according to Moravec et al. (1998) those are the main characters to allocate them as part of the superfamily Habronematoidea. Additionally, the nematode larva also has an oral opening dorsoventrally elongated, pseudolabia reduced to small appendage, and four cephalic papillae at the base of the pseudolabia. These features suggest that it may belong to the family Cystidicolidae, as proposed by Moravec et al. (1998) as diagnostic characteristics of this family. However, we could not perform a more accurate identification due to the absence of characters that only appear in the adult stage.

The superfamily Habronematoidea includes heterogeneous parasites found in all groups of vertebrates (Anderson, 2000).

Specifically, nematodes of the family Cystidicolidae have been found in Neotropical freshwater fishes, with some records from the Paraguay river basin (Moravec, 1998). The record of this larva could be considered an accidental infection in this amphibian host.

Discussion

Although the number of helminthological studies on South American amphibians has increased recently, only a small proportion of hosts have been studied (Campião et al., 2014, 2015; Chero et al., 2023; Draghi et al., 2020 a; González et al., 2021). In this study, we identified 17 helminth taxa; 14 of which represent new host records and 9 new geographic records (see Table 2). Some of these taxa have been previously reported in other amphibian host species and localities in the Neotropics, including larvae of *Centrorhynchus* and physalopterid nematodes, as well as adults of cosmocercid and molineid nematodes, all of which are generalist taxa (Campião et al., 2014, González et al., 2021). Additionally, Cosmocercidae family was the most diverse, with a total of five species. These observations are similar with those reported by Campião et al. (2014, 2015).

Table 2. Helminth parasites of amphibians from Dry and Humid Chaco ecoregions from Argentina

Hosts	<i>D. nanus</i>	<i>P. azureus</i>	<i>P. platensis</i>	<i>S. nasicus</i>	<i>T. typhoniuis</i>	<i>L. bufonius</i>	<i>L. latinasus</i>	<i>L. luclator</i>	<i>L. macrosternum</i>	<i>P. albonotatus</i>	<i>P. santafecinus</i>	<i>E. haroi</i>
ACANTHOCEPHALA												
Centrorhynchidae												
<i>Centrorhynchus</i> sp.	a *	a *									a *	
NEMATODA												
Rhabdiasidae												
<i>Rhabdias</i> sp.					a **							
<i>R. elegans</i>						b						
Strongyloididae												
<i>S. pereirai</i>				a **								
Molineidae												
<i>O. proencai</i>								a +				
<i>O. subauricularis</i>							c *			c *		
Pharyngodonidae												
<i>P. s. cubensis</i>					c **							
Anisakidae												
<i>Contracecum</i> sp.								a *				
Ascarididae												
<i>Porrocaecum</i> sp.					a *							
Cosmocercidae												
<i>A. hylambatis</i>							c					
<i>C. parva</i>								a				
<i>C. podicipinus</i>										c		
<i>C. phyllomedusae</i>		a +										
<i>S. formosula</i>							c *					
Kathlaniidae												
<i>F. macula</i>			a +					a	a +			
Physalopteridae												
<i>Physaloptera</i> sp.		a *						a *				b +, c
Larva aff. <i>Cystidicolidae</i>										c **		

Note. Sites of collection: a — Corrientes, Capital department, Corrientes province (Humid Chaco); b — Taco Pozo, Almirante Brown department, Chaco province (Dry Chaco); c: Ingeniero Juárez, Matacos department, Formosa province (Dry Chaco). * — First host report; + — First geographical report.

Table 3. Helminth parasites of *Leptodactylus luctator* collected in different provinces of Argentina

Hosts	Helminth taxa	Province	Reference
Acanthocephala			
Echinorhynchidae	<i>Pseudoacanthocephalus</i> . cf. <i>lutzi</i>	Buenos Aires	Draghi et al. (2020 a, 2020 b)
Digenea			
Diplodiscidae	<i>Catadiscus uruguayensis</i> Freitas & Lent, 1939	Buenos Aires	Suriano (1970), Ostrowski de Núñez (1979), Draghi et al. (2020 a, 2020 b)
	<i>Catadiscus corderoi</i> Mañé-Garzón, 1958	Misiones	Lunaschi & Drago (2010)
	<i>Catadiscus inopinatus</i> Freitas, 1941	Corrientes	Hamann et al. (2013 b)
Macroderoididae	<i>Rauschiella palmipedis</i> (Lutz, 1928) Sullivan, 1977	Buenos Aires	Suriano (1970); Draghi et al. (2020 a, 2020 b)
	<i>Rauschiella repandum</i> (Rudolphi, 1819)	Corrientes	Hamann et al. (2013 b)
	<i>Glythelmins bilialis</i> Suriano, 1968	Buenos Aires	Suriano (1968, 1970)
Gorgoderidae	<i>Gorgoderia australiensis</i> Johnston, 1912	Buenos Aires	Suriano (1965 a, 1970)
	<i>Gorgoderina parvicava</i> Travassos, 1920	Buenos Aires	Suriano (1965 b, 1970)
		Misiones	Lunaschi & Drago (2010)
		Corrientes	Hamann et al. (2013 b)
Plagiorchiidae	<i>Haematoloechus longiplexus</i> Stafford, 1902	Corrientes	Hamann et al. (2013 b)
	<i>Haematoloechus ozorioi</i> Freitas & Lent, 1939	Buenos Aires	Suriano (1970)
	<i>Choledocystus elegans</i> (Travassos, 1926) Ruiz 1949	Misiones	Lunaschi & Drago (2010)
	<i>Styphlodora</i> sp.	Corrientes	Hamann et al. (2013 b)
Derogenidae	<i>Halipegus dubius</i> Klein, 1905	Buenos Aires	Suriano (1970)
Diplostomidae	<i>Bursotrema tetracotyloides</i> Szidat, 1960	Buenos Aires	Szidat (1960)
		Corrientes	Hamann et al. (2013 b)

Continued Table 3

Hosts	Helminth taxa	Province	Reference
Nematoda			
Rhabdiasidae	<i>Rhabdias elegans</i> Gutiérrez, 1945	Buenos Aires	Draghi et al. (2020 a, 2020 b)
	<i>Rhabdias mucronata</i> Schuurmans Stekhoven, 1952	Corrientes	Schuurmans Stekhoven (1952)
Molineidae	<i>Oswaldocruzia proencai</i>	Corrientes	Present study
Anisakidae	<i>Contracaecum</i> sp.	Corrientes	Present study
Cosmocercidae	<i>Aplectana</i> sp.	Buenos Aires	Draghi et al. (2020 a)
	<i>A. hylambatis</i>	Buenos Aires	Draghi et al. (2020 a, 2020 b)
	<i>Cosmocerca parva</i>	Buenos Aires	Draghi et al. (2020 a, 2020 b)
		Corrientes	Present study
	<i>Cosmocercoides latrans</i> Draghi, Drago & Lunaschi, 2020	Buenos Aires	Draghi et al. (2020 a, 2020 b)
<i>Schrankiana</i> sp.	Buenos Aires	Draghi et al. (2020 a, 2020 b)	
Kathlaniidae	<i>Falcaustra mascula</i>	Not reported	Savazzini (1930)
		Corrientes	Present study
	<i>F. sanjuanensis</i> González, Sanabria & Quiroga, 2013	San Juan	Ramallo et al. (2016)
Physalopteridae	<i>Physaloptera</i> sp.	Corrientes	Present study

Among the hosts analyzed in this study, species such as *E. haroi* have few helminthological records. A cosmocercid, *Cosmocerca wichiorum* González, Hamann, Santos & Melo, 2023, was recently described from specimens of this microhylid collected from two areas of the Argentine Dry Chaco (González et al., 2023). Therefore, this represents the second record, adding a nematode taxon to its parasitic fauna. The limited knowledge of the helminth fauna of this species can be attributed to several factors, particularly the low number of publications and research efforts across all thematic areas, including parasitological studies, since its description in 2013 (Pereyra et al., 2023). Furthermore, its small size and fossorial habits likely contribute to this knowledge gap (Pereyra et al., 2013).

In contrast, *L. luctator*, a leptodactylid with a wide geographic distribution in the Neotropics, has the highest number of reports, which, according to Campião et al. (2014), may be attributable to the fact that this taxon forms a species complex. In Argentina, the parasitic fauna of this anuran has been stud-

ied in four provinces; the most significant number of records corresponds to Buenos Aires. For this host, nematodes and digeneans have been recorded throughout the territory, 12 and 14 taxa, respectively, along with a single species of acanthocephalans (Table 3).

This work expands the geographic distribution of three helminth species: *S. pereirai*, *P. senisfaciecaudus cubensis*, and *C. phyllomedusae*. These taxa are reported for the first time in Argentina; *S. pereirai* had previously been recorded only in Brazil, *P. senisfaciecaudus cubensis* only in Cuba, and *C. phyllomedusae* only in Peru and Paraguay (Barus & Coy Otero, 1969; Campião et al., 2014). In addition, *D. nanus* and *P. santafecinus* are reported as new hosts of *Centrorhynchus* sp.; *P. azureus* of *Centrorhynchus* sp. and *Physaloptera* sp.; *S. nasicus* of *S. pereirai*, *L. latinasus* of *O. subauricularis* and *S. formosula*; *P. albonotatus* of *O. subauricularis* and *C. podicipinus*; *T. typhonius* of *R. aff. pseudosphaerocephala*, *P. s. cubensis*, *Porrocaecum* sp., and *L. luctator* of *Contra-caecum* sp. and *Physaloptera* sp.

This work addresses one of the information gaps that Pereyra et al. (2023) identified in the study of Argentine amphibians. These authors define Pathologies as one of the 16 main research themes developed from 2010–2020. This theme ranked fifth in the studies conducted after Morphology, Ecotoxicology, Diversity and Distribution, and Ecology. However, it is worth noting that the authors included comprehensive and diverse topics such as malformations, fungi, and parasites within this theme. Undoubtedly, if only parasitic helminths were considered, the number of studies conducted on this theme would decrease despite the contributions made in the last 20 years.

Thus, although species such as *Leptodactylus latinasus*, *L. luctator*, and *L. macrosternum* have been the most studied in the last decade (Pereyra et al., 2023), studies to understand specifically their helminth fauna should continue over time and, also in new geographic areas from Argentina. A clear example is *L. luctator*, which, despite having helminth records from four provinces (Table 3), lacks helminthological studies in the rest of its extensive geographic distribution range in Argentina. Even more notable is the case of *L. latinasus*, a species distributed throughout 13 provinces of Argentina (Vaira et al., 2012), in which helminth studies have been conducted in only two localities, one in Corrientes province and the other in Chaco province (González & Hamann, 2015; Hamann et al., 2019).

This study contributes to filling the knowledge gaps on helminth parasites of Neotropical anurans, mainly in the Dry Chaco, one of the most threatened regions due to the synergistic combination of climatic and land use changes, principally in crops, pastures, and secondary scrub, which have the potential to produce the most dramatic impacts on land cover (Zak et al., 2008) and biodiversity loss (Nori et al., 2016). Nonetheless, only a minor fraction of anuran species has been examined in parasitological studies in the Neotropical region. Therefore, more studies will be necessary to elucidate this region's actual patterns of parasite biodiversity.

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