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FIRST REPORT OF NEMATODE PARASITES OF *PHYSALAELEMUS ALBONOTATUS*
(STEINDACHNER, 1864) (ANURA: LEIUPERIDAE) FROM CORRIENTES, ARGENTINA

PRIMER REPORTE DE NEMATODOS PARÁSITOS DE *PHYSALAELEMUS ALBONOTATUS*
(STEINDACHNER, 1864) (ANURA: LEIUPERIDAE) DE CORRIENTES, ARGENTINA

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Abstract

Thirty-four adults of *Physalaemus albonotatus* (20 males and 14 females) were collected near the city of Corrientes, Province of Corrientes, Argentina ($27^{\circ}28'S$ - $58^{\circ}50'W$) between January 2001 and September 2004. Adults of four nematode species, *Strongyloides* sp., *Oswaldocruzia* sp., *Cosmocerca podicipinus*, *Cosmocerca parva* and larvae of two other species, *Spiroxys* sp. and *Physaloptera* sp. were recovered from the digestive tract, lungs and gastric mucosa. We present morphologic characters (some with scanning electron microscopy), morphometric information and range extensions for these nematode species. This is the first report of nematode parasites of *P. albonotatus* from Argentina.

Key words: Amphibians - Argentina - Corrientes - nematode parasites - *Physalaemus albonotatus*.

Resumen

Treinta y cuatro adultos de *Physalaemus albonotatus* (20 machos y 14 hembras) fueron colectados en las proximidades de la ciudad de Corrientes, provincia de Corrientes, Argentina ($27^{\circ}28'S$ - $58^{\circ}50'W$) entre enero del 2001 y septiembre del 2004 para el estudio de su fauna nematológica. Un total de cuatro especies de nemátodos en estado adulto *Strongyloides* sp., *Oswaldocruzia* sp., *Cosmocerca podicipinus*, *Cosmocerca parva* y dos especies en estado larval, *Spiroxys* sp. y *Physaloptera* sp., fueron halladas en el tracto digestivo, pulmón y mucosa gástrica. En este estudio se presentan datos métricos y morfológicos de las especies halladas (algunos de ellos observados con microscopio electrónico de barrido). Éste es el primer reporte de nemátodos parásitos para *P. albonotatus* en Argentina.

Palabras clave: anfibios - Argentina - Corrientes - nemátodos parásitos, *Physalaemus albonotatus*.

INTRODUCTION

The nematode parasites of Argentinean amphibians were analyzed mainly for bufonids from northwestern (Ramallo *et al.*, 2007a, b, 2008) and for leptodactylids and bufonids from northeastern (González & Hamann, 2006a, b, 2007a, b, 2008, 2009; Hamann *et al.*, 2006a, b), while in the family Leiuperidae were made few studies of this topic. To our knowledge, in Argentina, the following leiuperids has been studied for nematode fauna: *Physalaemus biligonigerus* (Cope, 1861) from Cordoba province (Gutiérrez *et al.*, 2005) and, *Pseudopaludicola falcipes* (Hensel, 1867), *P. boliviensis* Parker, 1927 and *Physalaemus santafecinus* Barrio, 1965 from Corrientes province (Duré *et al.*, 2004; González & Hamann, 2004, 2009, 2010a). About the species *Physalaemus albonotatus* (Steindachner, 1864) this study is absent. Thus, the purpose of the present study is to report for the first time nematode species that found in adults specimens of *P. albonotatus* from Corrientes, Argentina.

This amphibian is found in Mato Grosso and Mato Grosso do Sul (Brazil), Paraguay, and the Chacoan regions of Bolivia and Argentina (Frost, 2011). In Argentina is common in natural environments but it can also be found in agricultural areas (Duré *et al.*, 2008). It can survive in both dry and moist substrata, i.e. near to the shore of temporary, semipermanent and permanent ponds, and also in the flooded grass. It has a diet dominated by formicids and isopterans and using an active foraging as strategy for prey capturing (Duré, 2004). There are no major threats to this species, is categorized as Least Concern by Aquino *et al.* (2004).

We described for the first time five genus of nematode parasites found in different organs of this amphibian host and presented new metrical and morphological data obtained with scanning electron microscopy.

MATERIAL AND METHODS

Thirty-four adults of *P. albonotatus* (20 males and 14 females) were collected near the city of Corrientes, Province of Corrientes in Argentina ($27^{\circ}28'S$ - $58^{\circ}50'W$) between January 2001 and September 2004. Frogs were hand-captured, mainly at night, using the sampling technique defined as “visual encounters survey” (Crump & Scott, 1994). Amphibians were transported live to the laboratory, killed in a chloroform (CHCl₃) solution. At necropsy, hosts were sexed (determined by examination of gonads and external nuptial features), and the alimentary canal, lungs, liver, kidneys, urinary bladder, musculature and integument examined for parasites by dissection. Nematodes were observed *in vivo*, counted and killed in hot distilled water and preserved in 70% ethyl alcohol, cleared in glycerine or lactophenol and examined as temporary mounts. Cysts were counted and isolated from host tissues. Larvae were removed from cysts using preparation needles and studied either *in vivo*. Some specimens were studied by scanning electron microscopy (SEM); these specimens were dehydrated in ethanol series, dried using the critical point technique, coated with gold, and examined with a JSM-5800 scanning electron microscope. Measurements are presented in micrometers (μm) unless otherwise indicated as mean values and standard deviation followed by range values in parenthesis. The systematic determination of the nematodes was carried out following the guidelines given by Anderson *et al.* (2009) and Gibbons (2010). Prevalence and mean intensity were calculated according to Bush *et al.* (1997).

The nematode specimens studied were deposited in the Helminthological Collection of the Centro de Ecología Aplicada del Litoral (CECOAL), Corrientes, Argentina. The present study has complied with all the regulations and ethical and legal considerations for the capture and use of animals established by the National Council of Scientific Research and Technical of Argentina

RESULTS

A total of 74 nematodes were collected from this leiuperid. These nematodes belonged to six species of the following families: Strongyloididae Chitwood and McIntosh, 1934: *Strongyloides* sp., Molineidae (Skrjabin and Schulz, 1937) Durette-Desset and Chabaud, 1977: *Oswaldocruzia* sp., Cosmocercidae Travassos, 1925: *Cosmocerca podicipinus* Baker and Vaucher, 1984 and *Cosmocerca parva* Travassos, 1925, Gnathostomatoidea Railliet, 1985: *Spiroxys* sp. and, Physalopteridae Leiper, 1908: *Physaloptera* sp. Each larva was spirally coiled inside the brownish capsule.

Strongyloides sp.

(Fig. 1A-B)

Prevalence, number of parasites: 2.9% (1 of 34 amphibians infected), 1.

Site of infection: large intestine.

Accession number: CECOAL 02113236 (1 gravid female).

Description: Based on 1 female specimen. Body slender, elongate, length 2.25mm, maximum width 55.0. Stoma simple, elongate, surrounded by six small lobes, each with one inconspicuous papilla. Esophagus filiform, 710.0 long. Nerve ring 170.0 from anterior end. Excretory pore not observed. Vulva with well developed lip situated at

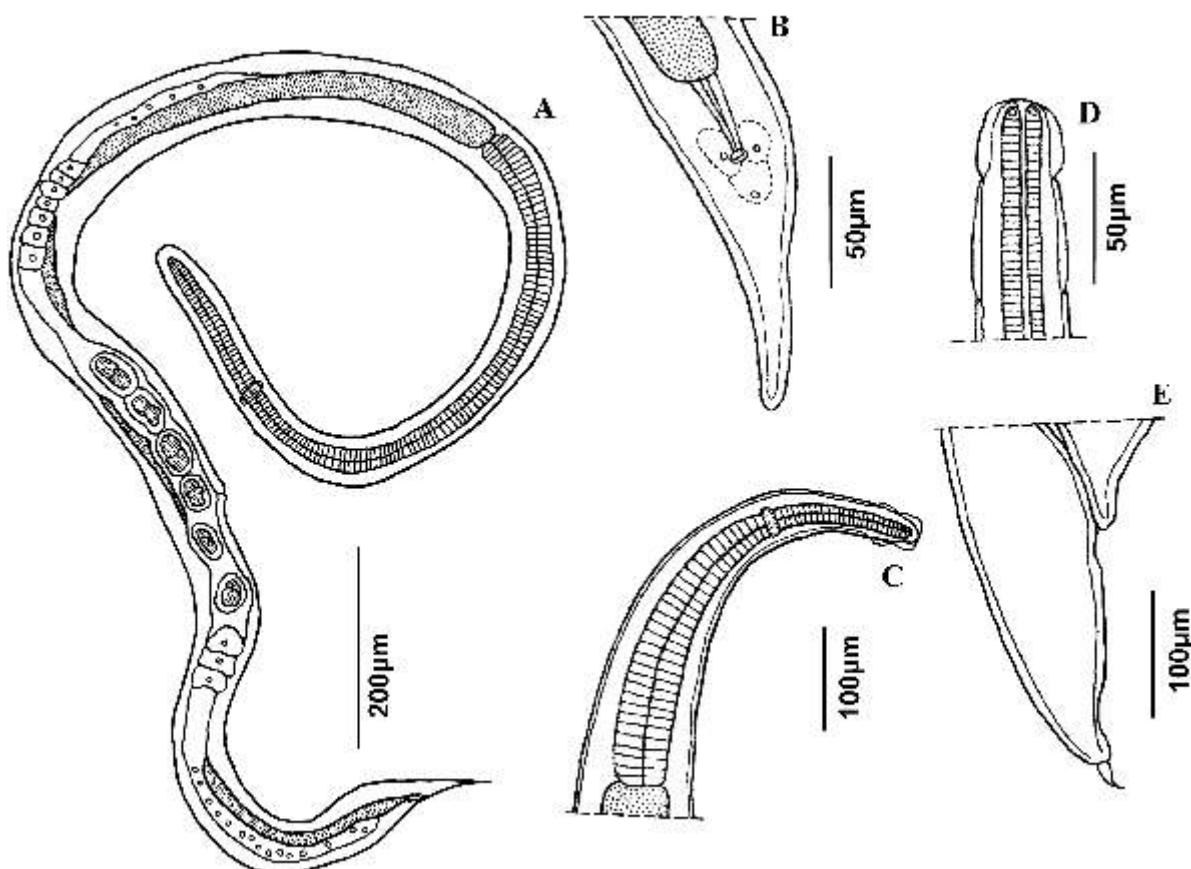


Figure 1A-E. Fig. 1A. *Strongyloides* sp. Female, total view. Fig. 1B. *Strongyloides* sp. Posterior end, ventral view. Fig. 1C. *Oswaldo cruzia* sp. Female, anterior end. Fig. 1D. *Oswaldo cruzia* sp. Detail of anterior end. Fig. 1E. *Oswaldo cruzia* sp. Posterior end, lateral view.

1.6mm from anterior end. Uteri opposed. Oval eggs, 50.0 in length x 34.0 wide. Anus 85.0 from posterior end; sharply pointed tail.

Remarks: The posterior end of these nematodes has been used as a diagnostic character for the species of this genus that parasitize anurans. Moravec and Kaiser (1995) have proposed two types: Type A: *Strongyloides* species that have a sharply pointed tail and, Type B: species that have a rounded tail. Specimen analyzed in this study might belong to the first type due to the morphology of the posterior end. In the present study, female nematode showed greater length (2.25mm) than female of *S. carinii* Pereira, 1935 (1.3-1.7mm) and *S. pereirai* Travassos, 1932 (1.56-1.91mm) and, the distance of anus to posterior end was greater (85.0) than those reported for these species (*S. carinii*: 60.0; *S. pereirai*: 64.0-72.0) analyzed by Pereira (1935) and Travassos (1932), respectively.

Geographical distribution: In amphibians from Neotropical Realm, species of this genus were found in leptodactylids and bufonids from Brazil (Travassos, 1932, 1933; Pereira, 1935; Luque *et al.*, 2005; Santos & Amato, 2010), in bufonids from Cuba (Pérez-Vigueras, 1942) and, in strabomantids from St. Vincent (Moravec & Kaiser, 1995). This is the first report of this genus for Argentinean amphibians.

Oswaldocruzia sp.

(Fig. 1C-E)

Prevalence, number of parasites: 2.9% (1 of 34 amphibians infected), 1.

Site of infection: small intestine.

Accession number: CECOAL 96040901 (1 immature female).

Description: Based on 1 female specimen. Body 9.0 mm x 150.0, with maximum width at level of midbody. Lateral alae undeveloped. Cephalic vesicle present with anterior swelling, total length of cephalic vesicle 87.4, rising approximately 6.9 above body wall in the widest first part. Claviform esophagus 540.0 length. Nerve ring, excretory pore and vulva 170.0, 282.0 and 6.37mm from

anterior end, respectively. Amphidelphic. Tail 207.0 in length, terminating in flexible filament approximately 11.0 in length. Female without eggs.

Remarks: The identification of the species of this genus is done by analyzing the caudal bursa of males, the morphology of the spicules and the constitution of synlophe (Ben Slimane *et al.*, 1996); because were not found males the specific identification was not possible.

Geographical distribution: *Oswaldocruzia* is a genus with a widely distribution in Neotropical Realm. Ben Slimane *et al.* (1996) reported species of this genus in leptodactylids from Chile, leptodactylids and bufonids from Brazil, leptodactylids from Paraguay, bufonids, hylids and leptodactylids from Guyana, bufonids from Venezuela, leptodactylids, bufonids and hylids from Ecuador and, in hylids, leptodactylids and bufonids from Cuba and Puerto Rico. In Argentina, previous reports in amphibians hosts were: *Oswaldocruzia mazzai* Travassos, 1935 in bufonids from Jujuy province (Travassos, 1935, 1937), *Oswaldocruzia* sp. in leptodactylids from Corrientes province (González & Hamann, 2006b) and, *O. proenca* Ben Slimane and Durette-Desset, 1995 in bufonids from Corrientes (González & Hamann, 2008) and Salta provinces (Ramallo *et al.*, 2007b). This is the first report of Argentinean leiuperid as host of genus *Oswaldocruzia*.

Cosmocerca podicipinus Baker and Vaucher, 1984

(Fig. 2A-B)

Prevalence, number of parasites: 73.5% (25 of 34 amphibians infected), 59.

Site of infection: lung, large intestine and small intestine.

Accession number: CECOAL 03074242 (1 male, 4 females); 02092915 (2 males, 4 females).

Description: Nematodes with conspicuous sexual dimorphism; body cuticle provided with transverse striations. Lateral alae present in both sexes. Mouth with 3 small v-shaped lips, dorsal lip with 2 sessile papillae, each ventrolateral lip with 1 ventral sessile papilla and 1 amphid; three

additional pairs of cephalic papillae. The anterior end of the esophagus presents three tooth-like projections covered with a thick cuticle, also called cuticular flap. Esophagus with short pharynx, cylindrical corpus, and posterior bulb containing valves. On the ventral surface of the preanal region of male there are two longitudinal rows of comb-like crests (plectanes). In this species there are 5 pairs of plectanes; the plectanes of each row are fused by sclerotized supports very marked (Fig. 2A-B). Somatic papillae numerous, in two ventral, two subventral, two dorsal and two subdorsal rows, extending from cervical region to posterior end. The metric characters of the males and females of these nematode species are present in Table 1.

Remarks: Some species of genus *Cosmocerca* have a fixed number of pairs of plectanes in the posterior end of males; *C. chilensis* Lent and Freitas, 1948 and *C. rara* Freitas and Vicente, 1966 have 6 pairs, *C. cruzi* Rodrigues and Fabio, 1970, *C. travassosi* Rodrigues and Fabio, 1970 and *C. podicipinus* have 5 pairs, *C. longispicula* Moravec and Kaiser, 1994 and *C. vrcibradici* Bursey and Goldberg, 1998 and *C. uruguayensis* Lent and Freitas, 1948 have 7 pairs. Other species have a variable number of pairs of these structures: *C. brasiliensis* Travassos, 1925 have 8-11 pairs, *C.*

paraguayensis Moravec and Kaiser, 1994 have 4-5 pairs and *C. parva* Travassos, 1925 have 4-7 pairs (Goldberg *et al.*, 2005). Thereon, specimens of *C. podicipinus* examined in this study show the same pattern as previous studies (5 pairs of plectanes); nevertheless, in some of immature specimens, the plectanes of each row do not fuse.

The metrical characteristic of *C. podicipinus* compared with original description realized by Baker and Vaucher (1984) with specimens collected in leptodactylids from Paraguay shows that males and females specimens analyzed from *P. albonotatus* are smaller than those studied by these authors (Baker & Vaucher, 1984: females: 2.86-7.33mm; males: 1.88-3.17mm vs. present study: females: 2.58-5.68mm; males: 1.35-2.95mm). In males, the length of spicules and gubernaculum are smaller than those found in Paraguayan specimens (Baker & Vaucher, 1984: spicules: 82-188; gubernaculum: 87-152 vs. present study: spicules: 57-97; gubernaculum: 62-141). Compared with specimens collected in a leiuperid sympatric host, *P. santafecinus* studied by González and Hamann (2010a), males analyzed in this study have total length of body, length of gubernaculum and length of spicules greater than males analyzed from *P. santafecinus* (*P.*

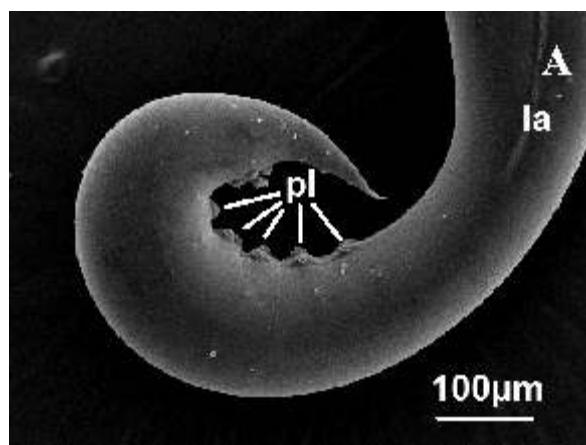


Figure 2A. *Cosmocerca podicipinus*. Posterior end of male, lateral view. pl: plectanes; la: lateral alae.

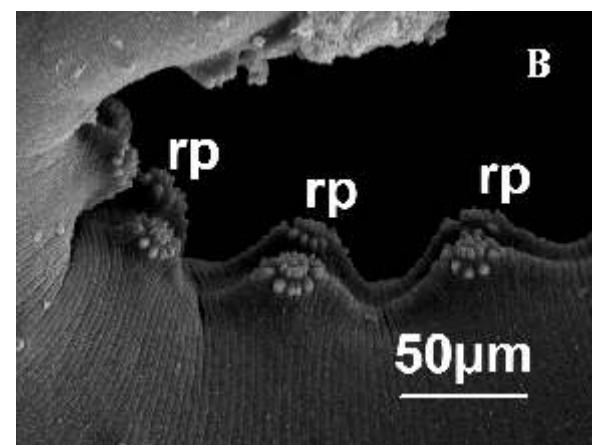


Figure 2B. *Cosmocerca podicipinus*. Posterior end of male; detail of plectanes. rp: rosette papillae.

Table 1. Morphological features and measurements of *Cosmocerca podicipinus* and *Cosmocerca parva* in *Physalaemus albonotatus*; measurements are presented in micrometers unless otherwise indicated as mean values and standard deviation followed by range values in parenthesis.

	<i>Cosmocerca podicipinus</i>		<i>Cosmocerca parva</i>	
	Females (n= 10)	Males (n= 10)	Females (n= 10)	Males (n= 10)
Total length	4.13±0.87mm (2.58–5.68)	2.16±0.56mm (1.35–2.95)	4.25±0.17mm (2.77–6.83)	1.52±0.25mm (1.15–1.72)
Width	220.5±33.1 (185–300)	201.8±57.3 (100–280)	272.5±61.30 (200–350)	158.5±19.6 (139–180)
Pharynx	35.6±7.7 (28–51)	26.6±4.1 (20–33)	42.2±5.9 (35–48)	30.0±1.4 (29–32)
	x	x	x	x
	34.3±6.3 (28–42)	18.1±3.4 (11–23)	36.9±4.8 (30–41)	21.5±1.7 (19–23)
Muscular esophagus	276.5±25.0 (234–306)	204.2±27.9 (172–270)	331.0±27.6 (294–360)	217.5±16.4 (198–232)
	x	x	x	x
	38.5±4.7 (31–46)	22.9±3.5 (17–30)	43.7±5.8 (37–49)	23.5±4.4 (21–30)
Bulb	84.6±11.4 (69–103)	55.6±7.4 (46–70)	100.0±13.1 (87–118)	57.0±7.8 (50–66)
	x	x	x	x
	91.3±11.1 (80–115)	55.1±7.7 (44–70)	101.7±16.1 (90–125)	57.7±6.4 (49–63)
Nerve ring *	209.5±27.0 (180–270)	173.0±33.6 (122–235)	214.0±25.7 (192–250)	147.5±6.4 (140–155)
Excretory pore *	336.6±49.8 (230–410)	265.2±46.0 (198–350)	354.0±26.0 (330–390)	208.5±34.3 (160–240)
Anus from posterior end	413.7±45.1 (350–510)	153.1±74.4 (103–360)	124.8±17.6 (102–140)	291.2±60.9 (225–370)
Gubernaculum	-	96.3±24.5 (62–141)	-	90.7±12.0 (78–107)
Spicules	-	85.3±10.7 (57–97)	-	109.7±15.8 (92–130)
Plectanes	-	5 pairs	-	4–6 pairs
Adanal papillae	-	3 pairs	-	3 pairs + 1
Vulva from anterior end	2.13±0.44mm (1.32–2.91)	-	2.25±0.20mm (2.0–2.5)	-
Eggs	95.4±12.4 (78–122)	-	101.2±12.3 (89–118)	-
	x	-	x	-
	61.0±7.7 (53–76)	-	58.2±7.7 (49–68)	-

* from anterior end.

santafecinus: total length: 1.3–2.3 mm; gubernaculum: 75–92; spicules: 69–89 vs. present study: total length: 1.35–2.95 mm; gubernaculum: 62–141; spicules: 57–97); females, however, have a total length less than those studied in *P. santafecinus* (*P. santafecinus*: 4.27–7.8 mm vs. present study: 2.58–5.68 mm).

Geographical distribution: In Neotropical Realm, the genus *Cosmocerca* has a widely distribution in amphibians hosts. *Cosmocerca podicipinus* was found in families Bufonidae, Aromobatidae, Strabomantidae and Leptodactylidae from Peru (Bursey *et al.*, 2001), in Leptodactylidae (Baker & Vaucher, 1984) and Hylidae from Paraguay (McAllister *et al.*, 2010b), Bufonidae and Dendrobatidae from Colombia (Goldberg & Bursey, 2003), Hylidae (Campião *et al.*, 2010; Goldberg *et al.*, 2007) and Leptodactylidae from Brazil (Campião *et al.*, 2009; Goldberg *et al.*, 2009), Dendrobatidae from Panama (Martinez & Maggenti, 1989), Eleutherodactylidae, Leptodactylidae, Hylidae (Goldberg & Bursey, 2002; Goldberg *et al.*, 2002a) and Ranidae (Goldberg & Bursey, 2002; Cabrera-Guzmán *et al.*, 2007) from Mexico, Bufonidae, Leptodactylidae, Microhylidae, Strabomantidae, Craugastoridae (Bursey & Brooks, 2010), Ranidae (Bursey & Goldberg, 2005, 2006, 2007; Bursey & Brooks, 2010), Brachycephalidae (Goldberg & Bursey, 2008a), and Hylidae (Goldberg & Bursey, 2008b) from Costa Rica. In Argentina, it was found in families Leiuperidae (González & Hamann, 2004, 2009a, 2010a), Leptodactylidae (González & Hamann, 2006b; Hamann *et al.*, 2006a, b; Schaefer *et al.*, 2006), Bufonidae (González & Hamann, 2006a, 2007a, b, 2008), Cycloramphidae (González & Hamann, 2009b) and Hylidae (González & Hamann, 2008, 2011; Hamann *et al.*, 2010). This is the first record of *C. podicipinus* in *P. albonotatus* from Argentina.

Cosmocerca parva Travassos, 1925

(Fig 3.A-B)

Prevalence, number of parasites: 8.8% (3 of 34 amphibians infected), 10.

Site of infection: large intestine.

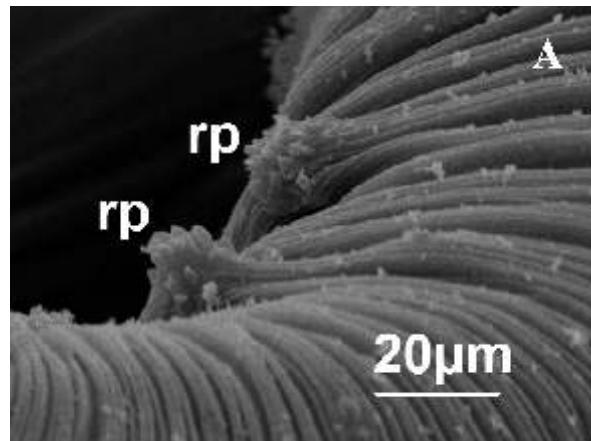


Figure 3A. *Cosmocerca parva*. Posterior end of male, lateral view. rp: rosette papillae.

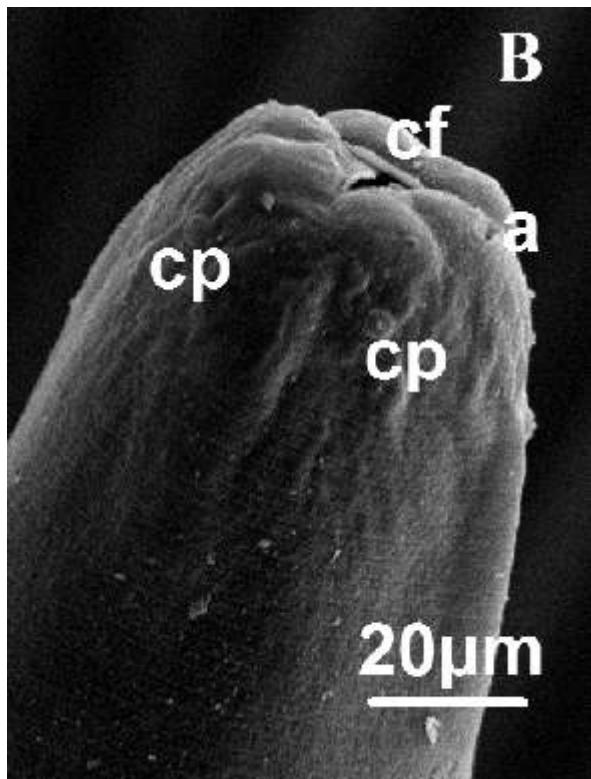


Figure 3B. *Cosmocerca parva*. Detail of anterior extremity of male. cp: cephalic papillae; a: amphid; cf: cuticular flap.

Accession number: CECOAL 02113235 (1 female), CECOAL 02103078 (1 female, 1 male).

Description: The Table 1 shows the metric characters of the males and females of *C. parva*. In this study, the number of plectanes and rosette papillae in the posterior end of male varied between 4 and 6 pairs, each rosette papillae with two complete circles of punctations directed perpendicular to the body surface; the plectanes of each row are not fused to each other as in the previous species (Fig. 3A). The anterior extremity of this species presents the same number and arrangement of cephalic papillae than *C. podicipinus* (Fig. 3B).

Remarks: For *C. parva* some studies show, in the same host, a fixed number of pairs of plectanes, in other hosts, the number of pairs of plectanes for this species was variable (see González & Hamann, 2010a). In this study we found males with new pattern of number of plectanes (4-6 pairs). On the other hand, adanals papillae present the same number and arrangement to those found in *P. santafecinus* (1 preanal pair, 1 lateral pair and 1 postanal pair); in addition, 1 unpaired papillae on the anterior lip of anus.

Studies realized with SEM in males of *C. parva* showed rosette papillae surrounded by 12-16 punctations in specimens collected in *R. granulosa* (Mordeglio & Digiani, 1998); surrounded by 12-15 interior punctations and 12-15 exterior punctations in specimens collected in *R. schneideri* (González & Hamann, 2008) and, rosette papillae surrounded by 10-11 interior punctations and 12-14 exterior punctations in specimens collected in *P. santafecinus* (González & Hamann, 2010a). Males of *C. parva* of this study shows 11 interior punctations and 12 exterior punctations in each one of rosette papillae.

Compared with original description realized by Travassos (1925) with specimens collected in Pernambuco State, Brazil, males of *C. parva* from *P. albonotatus* present a total length and the length of gubernaculum smaller than Brazilian specimens, but length of spicules is greater

(Travassos, 1925: total length: 3.5mm; gubernaculum: 120-140; spicules: 80 vs. present study: total length: 1.15-172mm; gubernaculum: 78-107; spicules: 92-130). Compared with specimens studied in a leiuperid sympatric host, *P. santafecinus*, by González and Hamann (2010a), the males of *C. parva* in this study are smaller and females are greater than those analyzed in *P. santafecinus* (*P. santafecinus*: males: 1.53-2.57mm; females: 3.43-5.92mm vs. present study: males: 1.15-1.72mm; females: 2.77-6.83mm). Also, the length of the spicules was higher in specimens collected from *P. albonotatus* (*P. santafecinus*: 57-98 vs. present study: 92-130).

Geographical distribution: In Neotropical Realm, *C. parva* was found in Brazil in the following families: Bufonidae (Santos & Amato, 2010), Leptodactylidae (Fabio, 1982), Leiuperidae and Hylolidae (Travassos, 1925); in Mexico from Ranidae (Paredes-Calderón *et al.*, 2004), in Guyana from Dendrobatidae and Hylidae (McAllister *et al.*, 2010a), in Paraguay from Leptodactylidae, Bufonidae (Baker & Vaucher, 1984, McAllister *et al.*, 2010b) and Hylidae (Masi Pallares & Maciel, 1974), in Peru from Bufonidae, Aromobatidae, Hylidae, Leiuperidae, Leptodactylidae, Eleutherodactylidae, Microhylidae (Bursey *et al.*, 2001), Strabomantidae and Dendrobatidae (McAllister *et al.*, 2010c), in Trinidad and Tobago from Leptodactylidae and Eleutherodactylidae (Goldberg *et al.*, 2002b), in Costa Rica from Brachycephalidae (Goldberg & Bursey, 2008a), Hylidae (Goldberg & Bursey, 2008b), Craugastoridae, Bufonidae, Eleutherodactylidae and Ranidae (Bursey & Brooks, 2010), in Colombia from Dendrobatidae (Sánchez *et al.*, 2010). In Argentina, it was found in the families Bufonidae (Mordeglio & Digiani, 1998; González & Hamann, 2006a, 2007a, b, 2008), Leptodactylidae (González & Hamann, 2006b; Hamann *et al.*, 2006a, b, Schaefer *et al.*, 2006), Cycloramphidae (González & Hamann, 2009b), Hylidae (González & Hamann, 2008, Hamann *et al.*, 2009, 2010) and Leiuperidae (González & Hamann, 2006b, 2010a). This is the first record of *C. parva* in *P. albonotatus* from Argentina.

Spiroxys sp. (Larvae)

(Fig. 4A-B)

Prevalence, number of parasites: 2.9% (1 of 34 amphibians infected), 1.

Site of infection: serous of stomach.

Accession number: CECOAL 10020901 (larvae).

Description: Based on 1 specimen. Small nematodes with fine cuticular transverse striations. Length of body 2.2 mm, maximum width 80.0. Cephalic end provided with two triangular lateral pseudolabia. Two cephalic papillae and one small amphid present on either side at level of base of pseudolabia. Walls of oral opening weakly sclerotized. Stoma weakly developed, very short. Esophagus starting at level of base of pseudolabia. Esophagus divided into narrow, anterior muscular

part (length 330.0) and broader posterior glandular part (length 750.0). Excretory pore not observed. Intestine brownish, straight. Tail conical, 90.0 long, with rounded tip. Elongated cyst, measures were 850.0 x 370.0.

Remarks: The identification of this larva is based mainly on the characteristic large lateral triangular pseudolabia. The principal differences compared with it found in *Pseudis paradoxa* from Corrientes, Argentina studied by González and Hamann (2010b) are the form of cyst (almost spherical in *P. paradoxa*, 380-400 x 350-375) and the smaller metrics characteristics (e.g. length of body: 1.97 mm; length of muscular esophagus: 100.0; length of glandular esophagus: 620.0; length of tail: 82.0).

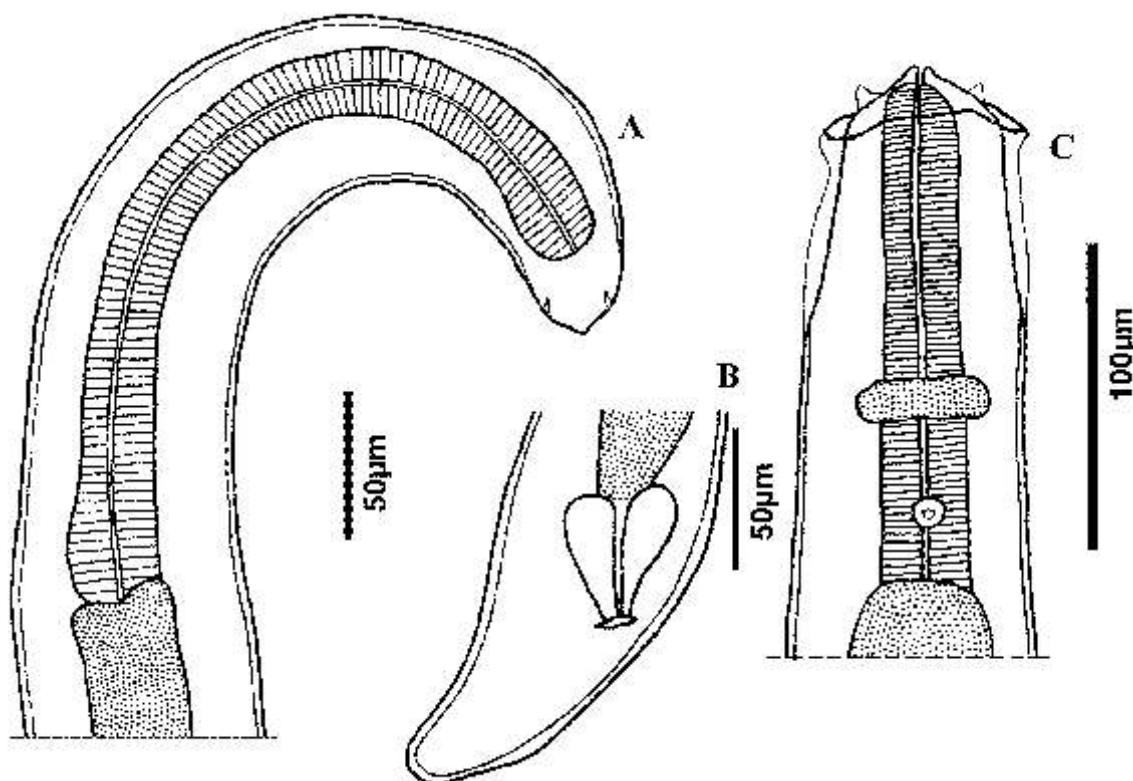


Figure 4A-C. Fig. 4A. *Spiroxys* sp. Anterior end, lateral view. Fig. 4B. *Spiroxys* sp. Posterior end, ventrolateral view. Fig. 4C. *Physaloptera* sp. Anterior end, ventral view.

Geographical distribution: In amphibian hosts of Neotropical Realm, the genus *Spiroxys* is previously reported in serous of stomach of hylid *P. paradoxa* from Argentina (González & Hamann, 2010b).

Physaloptera sp. (Larvae)

(Fig. 4C)

Prevalence, number of parasites: 2.9% (1 of 34 amphibians infected), 2.

Site of infection: gastric mucosa.

Accession number: CECOAL 06030612 (2 larvae).

Description: Based on 2 specimens. Larval body whitish, 2.56 ± 0.14 mm (2.55-2.57) long and 117.5 ± 3.5 (115.0-120.0) wide, with transversely annulated cuticle. Terminus of head with two lateral lips and cephalic collar formed by inflated cuticle. On either lip a sclerotized support and one terminal tooth present at upper margin. Each lip bearing two cephalic papillae and one amphid. Muscular esophagus 167.5 ± 3.5 (165.0-170.0) long by 32.0 ± 4.2 (29.0-35.0) wide; length of glandular esophagus 0.87 ± 0.03 mm (0.87-0.88) by 51.0 ± 1.4 (50.0-52.0) wide. Nerve ring encircling muscular esophagus at its posterior half, 115.0 ± 7.1 (110.0-120.0) from anterior end. Excretory pore near anterior end of glandular esophagus, 145.0 ± 7.1 (140.0-150.0) from anterior end of body. Tail conical, 110.0 ± 7.1 (105.0-115.0) long.

Remarks: Compared with larvae that found in other hosts of the same area, specimens analyzed from *P. albonotatus* showed the same morphological characteristics, but in terms of size, were smaller [total length in *P. santafecinus*: 3.13-3.7 mm (González & Hamann, 2010a); in *R. fernandezae*: 4.17 mm (González & Hamann, 2007a); in *R. granulosa*: 4.6-5.6 (González & Hamann, 2006a); in *R. schneideri*: 5.9 mm (González & Hamann, 2008)]. Also, the distance of the excretory pore and nerve ring from anterior end in specimens of this study are smaller than observed in *P. santafecinus* (nerve ring: 161.0-182.0, excretory pore: 168.0-204.0; González & Hamann, 2010a), in *R. schneideri* (nerve ring: 275.0, excretory pore: 360.0; González &

Hamann, 2008), in *R. granulosa* (nerve ring: 210.0-250.0, excretory pore: 240.0-340.0; González & Hamann, 2006a), in *R. fernandezae* (nerve ring: 162.0, excretory pore: 212.0; González & Hamann, 2007a).

Geographical distribution: In amphibians of Neotropical Realm larvae of *Physaloptera* were found in families Cycloramphidae (Boquimpani-Freitas et al., 2001), Leptodactylidae (Goldberg et al., 2009), Hylidae, Bufonidae and Leiuperidae (Vicente et al., 1990) from Brazil; in Bufonidae, Aromobatidae, Hylidae, Leiuperidae, Eleutherodactylidae, Leptodactylidae and Microhylidae (Bursey et al., 2001) from Peru, in Leptodactylidae (Goldberg et al., 2002b) from Trinidad and Tobago, Bufonidae (Galicia-Guerrero et al., 2000; Bursey & Goldberg, 2001; Goldberg et al., 2002a; Espinoza-Jiménez et al., 2007), Hylidae (Bursey & Goldberg, 2001; Goldberg et al., 2002a) and Ranidae (Bursey & Goldberg, 2001) from Mexico and, in Brachycephalidae (Goldberg & Bursey, 2008a) and Hylidae (Goldberg & Bursey, 2008b) from Costa Rica. In Argentina it was found in families Leiuperidae (Gutiérrez et al., 2005; González & Hamann, 2010a), Leptodactylidae (González & Hamann, 2006b), Hylidae and Bufonidae (González & Hamann, 2007a, 2008).

DISCUSSION

Of a total of nine species of the genus *Physalaemus* from Argentina, *P. albonotatus*, *P. biligonigerus*, *P. cuqui* Lobo, 1993, *P. cuvieri* Fitzinger, 1826, *P. fernandezae* (Müller, 1926), *P. gracilis* (Boulenger, 1883), *P. henselii* (Peters, 1872), *P. riograndensis* Milstead, 1960, *P. santafecinus* (Lavilla et al., 2000), the study of *P. albonotatus* represents the third report to the knowledge of the nematode fauna of this genus of amphibians (Gutiérrez et al., 2005; González & Hamann, 2010a).

Nematode parasites in the genus *Physalaemus* from Argentina include *Rhabdias* sp. and *Physaloptera* sp. in *P. biligonigerus* (Gutiérrez et

al., 2005), *C. podicipinus*, *C. parva*, *Aplectana hylambatis* (Baylis, 1927) Travassos, 1931 and *Physaloptera* sp. in *P. santafecinus* (González & Hamann, 2010a). Also, reports from Brazil (Vicente et al., 1990) included *Aplectana* sp., *A. lopesi* Silva, 1954, *A. membranosa* (Schneider, 1866) Miranda, 1924, *C. parva*, *Oxyascaris oxyascaris* Travassos, 1920 and *Physaloptera* sp. in *P. signifer* (Girard, 1853) and, *Aplectana* sp., *C. parva*, *O. oxyascaris* and *Physaloptera* sp. in *P. soaresi* Izecksohn, 1965. In the present study five genera of nematode parasites (*Strongyloides*, *Oswaldocruzia*, *Cosmocerca*, *Spiroxys* and *Physaloptera*) are reported for the first time in *P. albonotatus*. Additionally, the genus *Strongyloides* is reported for the first time in Argentinean amphibians.

The genera *Strongyloides*, *Oswaldocruzia* and *Cosmocerca* have a direct life cycle. The definitive host becomes infected by skin penetration of infective stage. Female nematodes expelled eggs that develop in the external environment into first-stage larvae and moult twice to the infective third stage. Migration to the lungs is apparently a necessary part of the development in species of genus *Cosmocerca* (Anderson, 2000). Nematodes of genera *Spiroxys* and *Physaloptera* have an indirect life cycle. They need to complete the cycle an intermediate host (arthropod) which is then consumed by the predaceous final host (Anderson, 2000). Adults stage of genus *Spiroxys* parasite stomach of different species of freshwater turtles.

Female nematodes expelled eggs with the turtle's faeces and the first-stage larvae develop in them. Then, it is ingested by a copepod and, in the haemocoel of this invertebrate, attains its third stage (Anderson, 2000). On the other hand, physalopterines are usually found firmly attached to the gastric mucosa with the aid of large dentate pseudalabia and a collarette which presses into the mucosa. In this way the larval parasite may persist for varying periods of time and be available to the predaceous final hosts (reptiles, birds, mammals) (Anderson, 2000). The presence of these larvae indicates that the amphibians could play a role in the transmission of these nematodes, serving as

paratenic host.

This study is the first report of nematode parasites of *P. albonotatus* from Argentina and some new morphological data are presented for all the species recorded, however due to the wide distribution of the host, future research should include references to nematode parasites of *P. albonotatus* collected in other localities.

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