



Research note

First report of helminth parasitizing *Trachycephalus typhonius* (Anura: Hylidae) from northeastern Argentina

Primer reporte de helmintos parasitando a Trachycephalus typhonius (Anura: Hylidae) del noreste de Argentina

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Abstract

Two species of nematodes, *Rhabdias* cf. *elegans* (Rhabdiasidae) and *Aplectana hylambatis* (Cosmocercidae), were recovered from the lungs and intestine of *Trachycephalus typhonius* (Anura: Hylidae) captured in Formosa Province, Argentina. Morphological and morphometric data of both species are provided, and previous records of *Rhabdias* spp. in the Neotropical region are summarized. The finding of *Rhabdias* cf. *elegans* constitutes the first record of this genus in the Formosa Province, and the finding of *A. hylambatis* represents the first record of this species as a parasite of hylid frogs, and the first report in Formosa Province. *Trachycephalus typhonius* represents a new host for both parasites.

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Keywords: *Rhabdias* cf. *elegans*; *Aplectana hylambatis*; Veined tree frog; Formosa Province

Resumen

Dos especies de nematodos, *Rhabdias* cf. *elegans* (Rhabdiasidae) y *Aplectana hylambatis* (Cosmocercidae), fueron recuperados de los pulmones e intestino de *Trachycephalus typhonius* (Anura: Hylidae) capturados en la provincia de Formosa, Argentina. Se proveen datos morfológicos y morfométricos de ambas especies y se resumen los registros previos de *Rhabdias* spp. en la región neotropical. El hallazgo de *Rhabdias* cf. *elegans* constituye el primer registro del género *Rhabdias* en la provincia de Formosa y el de *A. hylambatis* representa el primer registro de esta especie en hílidos, y el primer reporte en la provincia de Formosa. *Trachycephalus typhonius* es un nuevo hospedero para ambos parásitos.

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Palabras clave: *Rhabdias* cf. *elegans*; *Aplectana hylambatis*; Rana lechera común; Provincia de Formosa

The veined tree frog or common milk frog, *Trachycephalus typhonius* (Linnaeus, 1758) (Anura: Hylidae), is a very wide-spread frog from the lowlands of tropical Mexico, Central America (to 2,500 m asl in Guatemala), to Amazon Basin of Colombia, Ecuador, Peru, Venezuela, and Brazil, Guianas, and south to Brazil, Paraguay and northern Argentina (Frost, 2014; Soares, Iop, & Santos, 2012). In Argentina, this anuran species is distributed in Corrientes, Chaco, Entre Ríos, Formosa, Jujuy, Misiones, Salta, Santiago del Estero and Santa Fe Provinces (SIB, 2014). The veined tree frog is a nocturnal amphibian typ-

ically found perching on tree branches and vegetation while foraging at night. It preys on spiders, insects and bats, having a generalist diet (Duré & Kehr, 2006).

The helminth fauna of *T. typhonius* has been poorly studied, so far 6 species of helminths have been recorded parasitizing this species (cited as *Phrynohyas venulosa* and *Trachycephalus venulosus*): *Polystoma lopezromani* Combes and Laurent, 1979 (Monogenea: Polystomatidae) from Argentina (Combes & Laurent, 1979); *Batracholandros spectatus* (Freitas and Ibáñez, 1962) Freitas and Ibáñez, 1965 (Nematoda: Pharyngodonidae), and larvae of *Physaloptera* sp. (Nematoda: Physalopteridae) from Peru (Burse, Goldberg, & Parmalee, 2001); and *Parapharyngodon duniae* Bursey and Brooks, 2004 (Nematoda: Pharyngodonidae),

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Aplectana incerta Caballero, 1949 and *Aplectana itzocanensis* Bravo-Hollis, 1943 (Nematoda: Cosmocercidae) from Costa Rica (Burse & Brooks, 2004, 2010). The aim of this paper is to increase the knowledge of the diversity of helminth parasites from anurans from northeastern Argentina.

On September 17th 2012, 2 specimens of *T. typhonius* were collected at La Marcela farm in Pirané, Formosa Province, Argentina (26°17'35" S, 59°08'38" W), with authorization of Ministerio de la Producción y Ambiente, Dirección de Fauna y Parques of Formosa Province. The frogs were dissected after an overdose with 20% benzocaine and all organs were examined using a Stemi 2000-C Zeiss stereoscope. The collected nematodes were counted and fixed in 5% formaline and cleared in Aman's lactophenol for light microscopic investigation, using a Standard 25 Zeiss optical microscope. For their identification were used taxonomic descriptions taken from specific literature (Anderson & Bain, 1982; Baker & Vaucher, 1986; González, Quiroga, Moreno, & Sanabria, 2013; Gutiérrez, 1945). All measurements are given in micrometres (µm) unless otherwise stated, as the range followed by the mean and standard deviation in parentheses. The nematodes were deposited in the Helminthological Collection of the Museo de La Plata (MLP), and the hosts in the Herpetological Collection of the Museo de La Plata, La Plata, Argentina (accession numbers: MLP A5642, MLP A5643).

Rhabdiasidae Railliet, 1915

Rhabdias Stiles and Hassall, 1905

Rhabdias cf. *elegans* Gutiérrez, 1945 (Fig. 1)

Host: *Trachycephalus typhonius* (L.) (Anura: Hylidae).

New host record.

Site of infection: lungs.

Prevalence and intensity of infection: 50% (1 of 2); 2.

Voucher specimens deposited: MLP-He 6792.

Description

(Based on 2 hermaphrodite gravid specimens). Body cylindrical. Body length 5.9-6.4 (6.1±0.38) mm, maximum width 0.43-0.46 (0.44±0.02) mm. Anterior end rounded, posterior end conical. Body cuticle swollen, with irregular folds. Oral opening narrow, without labial structures. Buccal capsule infundibuliform, with sclerotized walls, 12-19 (15.6±5.3) deep, 16.5-19 (17.7±1.8) wide. Esophagus small, cylindrical, with a posterior dilatation. Esophagus length, 369-391 (380±15.71), 6-6.2 (6.1% of the body length). Esophagus width 45-57 (51±8.32) at anterior region. Posterior bulb 67-114 (90±33.48) wide. Inconspicuous nerve ring at 166.6 from the anterior region of the body (2.8% of the body length). Inconspicuous excretory duct. Intestine wide and filled with black and brown contents. Vulva near to middle body, slightly post-equatorial, distance from the anterior region of the body to vulva 3.3 mm (55% of body length). Genital system amphidelphic. Tail short, conical, 328.4-348 (338±13.66) long, representing 5.3-5.43 (5.5)% of body length. Eggs numerous, oval, thin-walled, in different stages of development and containing developed larvae near vulva, 101-110 (106±6.66) long by 49-66 (57±11.8) wide.

Remarks

Adult nematodes in the family Rhabdiasidae are common lung parasites of amphibians and reptiles, and the infection occur by skin penetration (Anderson, 2000), orally, or potentially via transport hosts (Baker, 1979). So far, of the approximately 90 species of this genus distributed worldwide (Tkach, Kuzmin, & Snyder, 2014), 21 species have been reported in the Neotropical Region (Table 1), and 5 of them in amphibians from Argentina: *Rhabdias fülleborni* Travassos, 1926 in *Rhinella schneideri* (Werner) from Corrientes Province; *Rhabdias elegans* Gutiérrez, 1945 in *Rhinella arenarum* (Hensel) from Buenos Aires and Salta Provinces and in *R. schneideri*, *Leptodactylus bufonius* Boulenger and *Odontophrynus americanus* (Duméril and Bibron) from Corrientes Province; *Rhabdias mucronata* Schuurmans Stekhoven, 1952 in *Leptodactylus latrans* (Steffen) from Corrientes Province; *Rhabdias truncata* Schuurmans Stekhoven, 1952 in *Telmatobius schreiteri* Vellard from Tucumán Province and *Rhabdias* aff. *sphaerocephala* in *Rhinella fernandezae* (Gallardo) from Corrientes Province. Moreover, specimens of *Rhabdias* sp. in *Leptodactylus chaquensis* Ceí and *Physalaemus biligonigerus* (Cope) were reported from Corrientes and Córdoba Provinces, respectively (González & Hamann, 2007, 2008, 2009; Gutiérrez, 1945; Gutiérrez, Attademo, Guerrero, Peltzer, & Lajmanovich, 2005; Kloss, 1971; Hamann, Kehr, & González, 2012, 2013; Schuurmans Stekhoven, 1952; Sueldo & Ramírez, 1976).

The specimens here studied are closely related to the original description of *R. elegans* when considering the slightly post-equatorial position of the vulva (3.3 vs. 2.4-4.4 mm from anterior region of the body), the body length (5.9-6.4 vs. 4.5-9.5 mm), the absence of lip structures in the anterior end, the cylindrical esophagus (369-391 vs. 314-490 long) and the eggs size (101-110 × 49-66 vs. 91-112 × 52-59). Given that have been found only 2 specimens and that has been stated by several researchers that the conservative morphology of the genus *Rhabdias* (uniform shape and size) and absence of males in the parasitic generation can result in the incorrect identification of some species (Kuzmin, Tkach, & Snyder, 2003; Martínez-Salazar, Pérez-Ponce de León, & Parra-Olea, 2009) more specimens will be necessary for a proper morphological and molecular taxonomic determination.

Cosmocercidae Railliet, 1916

Aplectana Railliet and Henry, 1916

Aplectana hylambatis (Baylis, 1927) Travassos, 1931

(Figs. 2, 3, and 4)

Host: *Trachycephalus typhonius* (L.) (Anura: Hylidae).

New host record.

Site of infection: small and large intestine.

Prevalence and intensity of infection: 50% (1 of 2); 48.

Voucher specimens deposited: MLP-He 6793.

Description

(Based on 15 specimens). Small, slender nematodes. Cuticle bearing transverse striations, uniform along the body. Triangular mouth surrounded by 3 lips: 1 dorsal with 2 cephalic papil-



Figures 1-4. Nematodes from *Trachycephalus typhonius*. 1. *Rhabdias* cf. *elegans*, hermaphrodite gravid specimen. Scale bar= 500 µm. 2. *Aplectana hylambatis*, female, entire worm. Scale bar= 200 µm. 3. *Aplectana hylambatis*, male, posterior extremity. Scale bar= 50 µm. 4. *Aplectana hylambatis*, male, enlarged view of the distal articulation of spicules. Scale bar= 50 µm. Abbreviations: da, distal articulation of the spicules; g, gubernaculum; po, postcloacal papillae; pr, precloacal papillae; s, spicules; v, vulva.

lae and 2 ventral with 1 cephalic papilla each. Two amphids. Esophageal bulb well distinct and with chitinous valves. Excretory pore pre-bulbar.

Female

(Based on 10 gravid specimens). Body 2.7-3.6 (3.2±0.28) mm long by 210-280 (244±21.7) wide. Pharynx muscular, 36-60 (45±7.85) long by 26-33 (30±2.11) wide; esophagus muscular, 386-467 (424±74.9) long; valvulated esophageal bulb 95-170 (108±22.35) long by 100-124 (110±6.9) wide. Nerve ring and excretory pore at 131-241 (210±55.15) and 238-483 (230±113.15) from anterior extremity, respectively. Prodelphic uterus. Vulva postequatorial at 1.9-2.3 (2.1±0.16) mm from anterior extremi-

ty, with scalloped edges. Eggs thin shelled, 75-91 long by 38-46 (81 × 53) wide. Tail 214-243 (230±10.88) long.

Male

(Based on 5 specimens). Body 2-2.4 (2.2±0.17) mm long by 120-220 (156±34.18) wide. Pharynx muscular, 36-45 (41±3.27) long by 18-24 (23±2.4) wide; esophagus muscular 357-448 (408±33.16) long; valvulated esophageal bulb 71-95 (83±12) long by 79-102 (87±10.04) wide. Nerve ring and excretory pore at 167 and 362-410 (362±23) from anterior extremity, respectively. Spicules long, equal, 296-328 (311±13.6) with a distal articulation that represents, approximately, 1/8 of its total length. Gubernaculum 68-88 (76±8.8) long. Caudal papillae distributed as follow: 3-4

Table 1
List of host species for Neotropical *Rhabdias* spp.

Host group	Host species	Species of <i>Rhabdias</i>	Country	Reference
Amphibia				
Bufonidae	<i>Anaxyrus kelloggi</i> (Taylor)	<i>R. americanus</i> Baker, 1978	Mexico	Goldberg & Bursey, 2002
	<i>Incilius occidentalis</i> (Camerano)	<i>R. kuzmini</i> Martínez-Salazar & León-Règagnon, 2007	Mexico	Martínez-Salazar & León-Règagnon, 2007
	<i>Incilius marmoratus</i> Wiegmann	<i>R. fülleborni</i>	Mexico	Galicia-Guerrero, Bursey, Goldberg, & Salgado-Maldonado, 2000
	<i>Incilius mazatlanensis</i> (Taylor)	<i>R. americanus</i>	Mexico	Goldberg & Bursey, 2002
	<i>Incilius nebulifer</i> (Girard)	<i>R. fülleborni</i>	Mexico	Galicia-Guerrero et al., 2000
	<i>Rhinella arenarum</i>	<i>R. elegans</i>	Argentina	Gutiérrez, 1945
			Paraguay	Kloss, 1971
			Uruguay	Kloss, 1971
			Brazil	Travassos, 1926
	<i>Rhinella bergi</i> (Céspedes)	<i>Rhabdias</i> sp.	Argentina	González & Hamann, 2007
	<i>Rhinella crucifer</i> (Wied)	<i>R. hermafrodita</i> Kloss, 1971	Brazil	Kloss, 1974
	<i>Rhinella fernandezae</i>	<i>R. aff. sphaerocephala</i>	Argentina	Hamann et al., 2013
	<i>Rhinella icterica</i> (Spix)	<i>R. fülleborni</i>	Brazil	Travassos, 1926; Kloss, 1971; Vicente, Rodrigues, Gomes, & Pinto, 1991; Luque, Martins, & Tavares, 2005; Pinhão, Wunderlich, Anjos, & Silva, 2009
	<i>Rhinella marina</i> L.	<i>R. alabialis</i> Kuzmin, Tkach and Books, 2007	Costa Rica	Kuzmin, Tkach, & Books, 2007
		<i>R. americanus</i>	Mexico	Espinoza-Jiménez, García-Prieto, Osorio-Sarabia, & León-Règagnon, 2007
		<i>R. fülleborni</i>	Brazil	Kloss, 1971; Kloss, 1974
			Mexico	Galicia-Guerrero et al., 2000; Goldberg, Bursey, Salgado-Maldonado, Baéz, & Cañeda, 2002; Espinoza-Jiménez et al., 2007
		<i>R. paraensis</i> Santos, Melo, Nascimento, Nascimento, Giese & Furtado, 2011	Brazil	Santos et al., 2011
		<i>R. pseudosphaerocephala</i> Kuzmin, Tkach & Brooks, 2007 (as <i>Rhabdias sphaerocephala</i> Goodey, 1924)	Brazil	Kloss, 1971, 1974
		<i>Rhabdias</i> sp.	Bermuda Island	Kloss, 1974
		Mexico	Galicia-Guerrero et al., 2000; Guillén-Hernández, 1992	
		Nicaragua	Kuzmin et al., 2007	
		Costa Rica	Kuzmin et al., 2007	
	<i>Rhinella margaritifera</i> (Laurenti)	<i>R. androgyna</i> Kloss, 1971	Brazil	Kloss, 1971
	<i>Rhinella rubescens</i> (Lutz)	<i>R. elegans</i>	Brazil	Kloss, 1971
	<i>Rhinella schneideri</i>	<i>R. fülleborni</i>	Brazil	Kloss, 1971; Vicente et al., 1991
		<i>R. elegans</i>	Argentina	Kloss, 1971
Craugastoridae	<i>Craugastor occidentalis</i> (Taylor)	<i>R. manatlanensis</i> Martínez-Salazar, 2008	Mexico	Martínez-Salazar, 2008
Cycloramphidae	<i>Thoropa miliaris</i> (Spix)	<i>R. fülleborni</i>	Brazil	Travassos, 1926
Hylidae	<i>Dendropsophus microcephalus</i> (Cope)	<i>Rhabdias</i> sp. [= <i>R. tobagoensis</i> of Goldberg, Bursey, Salgado-Maldonado et al. (2002)]	Mexico	Martínez-Salazar et al., 2009
	<i>Hypsiboas albopunctatus</i> (Spix)	<i>Rhabdias</i> sp.	Brazil	Holmes, Bocchiglieri, Araújo, & Silva, 2008
	<i>Pseudacris hypochondriaca</i> (Hallowell)	<i>R. peninsularis</i> Martínez-Salazar, Falcón-Ordaz, González-Bernal, Parra-Olea & Pérez Ponce de León, 2013	Mexico	Martínez-Salazar, Falcón-Ordaz, González-Bernal, Parra-Olea, & Pérez Ponce de León, 2013
	<i>Pseudacris regilla</i> (Baird and Girard)	<i>R. peninsularis</i> (as <i>R. ranae</i>)	Mexico	Goldberg, Bursey, & Gergus, 2001
	<i>Pseudis platensis</i> Gallardo	<i>Rhabdias</i> sp.	Brazil	Campiã, da Silva, & Ferreira, 2010
	<i>Scinax staufferi</i> (Cope)	<i>Rhabdias</i> sp.	Mexico	Martínez-Salazar et al., 2009
	<i>Smilisca baudinii</i> Duméril and Bibron	<i>R. americanus</i>	Mexico	Goldberg & Bursey, 2002
		<i>Rhabdias</i> sp.	Mexico	Guillén-Hernández, 1992
	<i>Smilisca cyanosticta</i> (Smith)	<i>R. fülleborni</i>	Mexico	Goldberg et al., 2002
	<i>Trachycephalus typhonius</i>	<i>Rhabdias</i> cf. <i>elegans</i>	Argentina	Present study
Leptodactylidae	<i>Leptodactylus chaquensis</i>	<i>Rhabdias</i> sp.	Brazil	Vicente et al., 1991
		<i>Rhabdias</i> sp.	Argentina	Hamann, Kehr, & González, 2006
	<i>Leptodactylus bufonius</i>	<i>R. elegans</i>	Argentina	Hamann et al., 2012

Table 1
List of host species for Neotropical *Rhabdias* spp. (Cont.)

Host group	Host species	Species of <i>Rhabdias</i>	Country	Reference
Leptodactylidae (cont.)	<i>Leptodactylus labyrinthicus</i> (Spix)	<i>R. fülleborni</i>	Brazil	Vicente et al., 1991
	<i>Leptodactylus latrans</i>	<i>R. mucronata</i>	Argentina	Schuermans-Stekhoven, 1952
	<i>Leptodactylus macrosternum</i> Miranda	<i>R. breviensis</i> Nascimento, Goncalves, Melo, Giese, Furtado and Santos, 2013	Brazil	Nascimento et al., 2013
	<i>Leptodactylus melanonotus</i> (Hallowell)	<i>R. elegans</i>	Mexico	Goldberg et al., 2002
		<i>R. elegans</i>	Mexico	Martínez-Salazar & León-Règagnon, 2007
		<i>R. ranae</i> Walton, 1929	Mexico	Goldberg & Bursey, 2002
		<i>Rhabdias</i> sp.	Mexico	Mata-López, León-Règagnon, & García-Prieto, 2013
	<i>Leptodactylus podicipinus</i> (Cope)	<i>Rhabdias</i> sp.	Brazil	Campião, da Silva, & Ferreira, 2009
	<i>Leptodactylus syphax</i> Bokermann	<i>Rhabdias</i> sp.	Brazil	Vicente et al., 1991
	<i>Physalaemus biligonigerus</i>	<i>Rhabdias</i> sp.	Argentina	Gutiérrez et al., 2005
	<i>Pristimantis terraebolivaris</i> (Rivero)	<i>R. tobagoensis</i> Moravec & Kaiser, 1995	Tobago	Moravec & Kaiser, 1995
	Odontophrynidae Ranidae	<i>Odontophrynus americanus</i>	<i>R. elegans</i>	Argentina
<i>Lithobates berlandieri</i> (Baird)		<i>R. fülleborni</i>	Mexico	León-Règagnon, Martínez-Salazar, Lazcano-Villarreal, & Rosas-Valdéz, 2005
		<i>R. savagei</i> Bursey and Goldberg, 2005	Mexico	León-Règagnon et al., 2005
		<i>Rhabdias</i> sp.	Mexico	Guillén-Hernández, 1992
<i>Lithobates forreri</i> (Boulenger)		<i>R. savagei</i>	Costa Rica	Bursey & Goldberg, 2005
<i>Lithobates</i> cf. <i>forreri</i>		<i>R. pseudosphaerocephala</i>	Mexico	Cabrera-Guzmán, León-Règagnon, & García-Prieto, 2007
<i>Lithobates magnaocularis</i> (Frost and Bagnara)		<i>R. ranae</i>	Mexico	Goldberg & Bursey, 2002
<i>Lithobates tarahumarae</i> (Boulenger)		<i>R. ranae</i>	Mexico	Bursey & Goldberg, 2001
<i>Lithobates vaillanti</i> (Brocchi)		<i>R. fülleborni</i>	Mexico	Goldberg et al., 2002
		<i>R. pseudosphaerocephala</i>	Mexico	Paredes-Calderón, León Règagnon, & García-Prieto, 2004
		<i>Rhabdias</i> sp.	Mexico	Guillén-Hernández, 1992
		<i>R. pseudosphaerocephala</i>	Mexico	Martínez-Villarreal, 1969
Telmatobiidae	<i>Lithobates</i> sp.	<i>R. pseudosphaerocephala</i>	Mexico	Martínez-Villarreal, 1969
	<i>Telmatobius schreiteri</i>	<i>R. truncata</i>	Argentina	Schuermans-Stekhoven, 1952
Reptilia Anguidae Dactyloidae	<i>Elgaria paucicarinata</i> (Fitch)	<i>Rhabdias</i> sp.	Mexico	Goldberg & Bursey, 2004
	<i>Anolis frenatus</i> (Cope)	<i>R. anolis</i> Bursey, Goldberg & Telford, 2003	Panamá	Bursey, Goldberg, & Telford, 2003
	<i>Anolis megapholidotus</i> Smith	<i>R. leonae</i> Martínez-Salazar, 2006	Mexico	Martínez-Salazar, 2006
	<i>Anolis capito</i> Peters	<i>R. nicaraguensis</i> Bursey, Goldberg & Vitt, 2007	Nicaragua	Bursey, Goldberg, & Vitt, 2007

pairs of ventral precloacal papillae, 1 sublateral adcloacal pair, 3 pairs on anterior lip, 1 unpaired papilla on anterior lip, 2 pairs of ventrolateral postcloacal papillae in the median region of the tail and 1 pair next to the posterior end. Tail 121-214 (172±33.6) long.

Remarks

Considering the revision of the genus *Aplectana* performed by Baker (1980) and the posterior descriptions and synonymies (Baker & Vaucher, 1986; González et al., 2013), *A. hylambatis* shares with *Aplectana chamaeleonis* (Baylis, 1929) Travassos, 1931, the characteristic morphology of the spicules. However, the specimens here described are easily distinguished from *A. chamaeleonis* by the presence of 2 pairs of ventrolateral postcloacal papillae in the median region of the tail. In addition, the papillae disposition, the size of the spicules and gubernaculum in the male and the postequatorial vulva with scalloped edges in the females allowed the specific determination.

In general, the specimens here described are similar in size to those described by González and Hamann (2010) parasitizing *Physalaemus santafecinus* from Corrientes Province, but slightly smaller than the specimens described by Gutiérrez (1945) in *R. arenarum* from Buenos Aires Province, those described by Baker (1980) in *Rhinella achalensis* from Córdoba Province and those described by González et al. (2013) in *R. arenarum* from San Juan Province, Argentina.

Aplectana hylambatis is a generalist species parasitizing a wide range of anurans hosts. In Argentina, it was found parasitizing 2 families of frogs: Bufonidae and Leptodactylidae (subfamilies Leptodactylinae and Leiuperinae) from Buenos Aires, Córdoba, Corrientes, Salta and San Juan Provinces (González et al., 2013).

This note represents the first record of *A. hylambatis* as a parasite of hylid frogs from Argentina, and the findings of *A. hylambatis* and *R. cf. elegans* constitute the first records of both genera in the Formosa Province. *Trachycephalus typhoni* represents a new host for both parasites.

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