

## Research Note

# Nematodes of the *Rhinella granulosa* Spix, 1824 (Anura: Bufonidae) from the Semiarid Northeastern Caatinga Region of Brazil

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**ABSTRACT:** We examined 31 specimens of *Rhinella granulosa* from the semiarid Caatinga region of northeastern Brazil for the presence of gastrointestinal nematodes. We found 7 nematode taxa: *Rhabdias androgyna*, *Raillietnema spectans*, *Aplectana membranosa*, *Oswaldocruzia* sp., *Physaloptera* sp., *Raillietnema* sp., and cosmocercidae larvae. The total prevalence was 25.8% and by mean intensity of infection ( $\pm$  SD) was  $50 \pm 7.36$ . The prevalence in males was 10% with an intensity of 4 while in females it was 42.85% with a mean intensity of  $42 \pm 8.55$ . The juveniles presented a prevalence of 14.28% and an intensity of 4. *Rhinella granulosa* represents a new host for the following nematodes: *Rhabdias androgyna*, *Raillietnema spectans*, and *Raillietnema* sp.

**KEY WORDS:** endoparasites, *Rhabdias androgyna*, parasitism in toads, *Raillietnema spectans*.

In the semiarid Caatinga region of northeastern Brazil, helminth investigations in anurans are still incipient, although an increasing number of studies have been completed over the last several years (Teles et al., 2014, 2015, 2016; Teles et al., 2017; Araujo Filho et al., 2015; Nascimento et al. 2012).

The granular or common lesser toad, *Rhinella granulosa* Spix, 1824, occurs from the east of Rio de Janeiro to the north of Brazil through Minas Gerais, Espírito Santo, Bahia, Piauí, Ceará, Alagoas, and Maranhão in all Atlantic Forest sites and east of Pernambuco and Rio Grande do Norte in the semiarid Caatinga (Frost, 2016). Most *Rhinella* frogs feed essentially on arthropods, mainly coleoptera and formicid hymenopterans (Sabagh and Carvalho-Silva, 2008). The helminth fauna of these toads is composed of acanthocephalans (Smales, 2007), nematodes, and

trematodes (Campião et al., 2014). The aim of the present study was to report the nematode faunal composition of *R. granulosa* in one area of the semiarid Caatinga region of Northeastern Brazil. Data are presented as means  $\pm$  SD.

We examined 31 *R. granulosa*, 10 males (mean snout–vent length [SVL] =  $20.54 \pm 3.5$  mm), 21 females (mean SVL =  $20 \pm 7.7$  mm), and 7 juveniles (mean SVL =  $2.57 \pm 1.5$  mm). Toads were collected manually in May 2011 at the Estação Ecológica de Aiuaba (ESEC-Aiuaba) ( $6.573476^{\circ}$ S;  $40.123564^{\circ}$ W) located in the Aiuaba municipality of the state of Ceará, Brazil. Immediately upon capture all specimens were given a lethal dose of 2% xylocaine, fixed with 10% formalin, and later preserved in 70% alcohol. The SVL was measured using a digital caliper (0.5-mm accuracy). We identified the sex of specimens by direct observation of their gonads. The digestive and respiratory tracts were examined for nematodes under a stereomicroscope. We identified the site of infection and counted all nematodes, which were then preserved in 70% alcohol in vials for subsequent taxonomic identification. Nematodes were cleared in Hoyer's medium (Everheart, 1957) and identified according to Anderson (2000) and Bursey et al. (1998). Specimens were later deposited in the parasitological collection of the Universidade Regional do Cariri (URCA-P), accession numbers (URCA-P: 509–516). The prevalence and intensity of infection were calculated according to Bush et al. (1997).

We found 50 specimens of nematodes infecting *R. granulosa*; 48 were found in the digestive tract and only 2 infected the lungs. We identified 7 taxa: *Rhabdias androgyna* Kloss, 1971, *Raillietnema*

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**Table 1. Abundance (A), Prevalence (P), Intensity (I), Range, and site of infection of *Rhinella granulosa* examined in this study by nematode species. The asterisk (\*) indicates a new species and a dash (–) indicates parasites were found in a single host.**

Nematode species	A	P	I	Range	Site of infection
<i>Aplectana membranosa</i>	28	6.45	14	(9–19)	Large intestines
Cosmocercidae (larva)	1	3.22	1	–	Large intestines
<i>Oswaldocruzia</i> sp.	9	9.67	3	(1–4)	Small intestines
<i>Physaloptera</i> sp.	5	6.45	2.5	(1–4)	Stomach
<i>Raillietnema spectans</i>	3	3.22	3	–	Large intestines
<i>Raillietnema</i> sp.*	2	3.22	2	–	Large intestines
<i>Rhabdias androgyna</i>	2	6.45	2	–	Lungs

*spectans* Gomes, 1964, *Aplectana membranosa* Miranda, 1924, *Oswaldocruzia* sp., *Physaloptera* sp., *Raillietnema* sp., and a cosmocercidae larva (Table 1). The total prevalence was 25.8% (8/31) and mean infection intensity was  $50 \pm 7.36$ . The prevalence in males was 10% (1/10, intensity 4) and in females 42.85% (6/14, mean intensity  $42 \pm 8.55$ ). One out of 7 juveniles was infected (prevalence = 14.28, intensity = 4). *Rhinella granulosa* represents a new host for the following nematodes: *Rhabdias androgyna*, *Raillietnema spectans*, and *Raillietnema* sp. All nematodes reported here correspond to new distribution records associated with this host in the semiarid Caatinga region. The nematodes *Rhabdias* Stiles & Hassal, 1905 are commonly found infecting the lungs of anurans and less commonly lizards, snakes, and salamanders (Baker, 1987; Martínez-Salazar and León-Règagnon, 2006; Ribeiro et al., 2012; Teles et al., 2014). Species of *Rhabdias* are distinguished on the basis of the buccal capsule and characteristics of the amphidelphic genital system. There are approximately 94 species belonging to this genus worldwide (Kuzmin et al., 2012).

In South America, there are 19 species of *Rhabdias* reported to infect the respiratory tracts of amphibians (Kuzmin et al., 2016). *Rhabdias androgyna*, at the moment, is recognized to infect the bufonid *Rhinella margaritifera* (= *Bufo typhonius*) in both Brazil and Guyana (Vicente et al., 1990; McAllister et al., 2010) and the odontophrynid, *Proceratophrys appendiculata*, in Brazil (Boquimpani-Freitas et al., 2001). The genus *Raillietnema* Travassos, 1927 is currently represented by 9 species in the Neotropical region: *R. baylisi* Walton, 1933; *R. brachyspiculatum* Bursey et al., 1998; *R. gubernaculatum* Freitas and Ibanez, 1965; *R. kritscheri* Moravec, Maldonado and Lope, 1993; *R. simples* Travassos, 1925; *R. mi-*

*nor* Freitas and Dobbin Jr., 1961; *R. lynchi* Bursey and Goldberg, 2006; *R. ibañezi* Cordova, 1998; and *R. spectans* Gomes, 1964, all of which infect lizards (Bursey et al., 1998) and, more frequently, anurans in South America (Vicente et al., 1991; Teles et al., 2015). To our knowledge, in Brazil the parasite *R. spectans* has been reported only for anurans. The anurans previously reported to be infected by this nematode were *R. crucifer*, *R. ictérica*, *R. schneideri*, *Leptodactylus latrans* (Vicente et al., 1991), and *Pleurodema diplolister* in the Caatinga (Teles et al., 2015). Specimens of the genus *Physaloptera* are generally found in the stomach of vertebrates such as amphibians, reptiles, mammals, birds, and rarely in fishes (Anderson, 2000). In Brazil, specimens of *Physaloptera* have been recorded from the following anuran families: Bufonidae (Gonçalves et al., 2002), Hylidae (Vicente et al., 1991), Leiuperidae (Fabio, 1982; Vicente et al., 1991), Leptodactylidae (Vicente et al., 1991), and Odontophrynidae (Klaion et al., 2011). Our specimens of *Physaloptera* were not identified to species level, as the sample contained only juveniles. Larvae belonging to the family Cosmocercidae can be found in the lungs and intestines of reptiles and anurans (Pinhão et al., 2009; Ávila and Silva, 2013) while in the present study, we found cosmocercid larvae infecting the stomach and intestines of our toads but were unable to identify them to species because of their immature nature. Nematodes of the *Oswaldocruzia* genus are known to infect anurans (Ben Slimane and Duret-Desset, 1996; Teles et al., 2016) and lizards (Oliveira et al., 2017) in the Neotropical region. At the present moment, approximately 90 species belonging to this genus are recognized (Ben Slimane e Duret-Desset, 1996), of which 13 have previously been recorded in anurans in South America (Campião et al., 2014). Infections caused by *Oswaldocruzia* species were recorded in the following families of anurans in South America: Bufonidae, Hylidae, Leiuperidae, and Leptodactylidae (Ben-Slimane and Durette-Desset 1996; Luque et al., 2005; Gonzalez and Hamann, 2006; Campião et al., 2014).

The parasite *Aplectana membranosa* is known to infect only anurans and is currently found in the following countries: Brazil (Campião et al., 2014), Ecuador (McAllister et al., 2010), Guyana (McAllister et al. 2010), and Uruguay (Lent and Freitas, 1948). The following anurans have been recorded as hosts of this parasite in Brazil: *Ischnochnema parva*, *Leptodactylus mystaceus*, *L. latrans*, *L. lineatus*, *L. pentadactylus*,

*L. vastus*, *Odontophrynus americana*, *Rhinella granulosa*, *R. icterica*, *R. marina*, *R. jimii*, and *R. schneideri*. *Aplectana membranosa* occurs in their hosts' rectum and large intestine (see Campião et al., 2014), as observed in the present study.

The parasitological indices reported in both the present and previous studies (Anderson, 2000; Pinhão et al., 2009; Campião et al., 2014) suggest that *Physaloptera* spp., *Rhabdias* spp., and *Oswaldocruzia* sp. are generalist parasites from anurans belonging to family Bufonidae. Our findings contribute to the increasing knowledge base concerning the parasitic fauna of Bufonidae in the Caatinga. Nevertheless, additional studies are important to understand the intricacies of parasite–host associations in such semiarid areas of the Neotropical region.

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