



New records of Helminths in Reptiles from five states of Brazil

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Abstract

Forty five specimens representing nine species of reptile (*Salvator merianae*, *Enyalius bilineatus*, *Amphisbaena alba*, *Xenopholis undulatus*, *Chironius fuscus*, *Helicops angulatus*, *Chironius flavolineatus*, *Erythrolamprus viridis* and *Crotalus durissus*) collected in five Brazilian states were examined for helminths. Twelve helminth species were found as follow: nine Nematoda (*Physaloptera tupinambae*, *Strongyluris oscari*, *Paracapillaria* sp., *Dracunculus brasiliensis*, *Physaloptera liophis*, *Serpentirhabias* sp. 1, *Serpentirhabias* sp. 2, *Serpentirhabias* sp. 3 and *Aplectana* sp.), one Cestoda (*Semenoviella amphisbaenia*), one Trematoda (*Paracotyletrema* sp.), and one Acantocephala (*Centrorhynchus* sp.). Ten new host records and seven new locality records were reported.

Keywords: parasite, snakes, lizards, amphisbaenidae, nematoda.

Novos registros de helmintos em répteis de cinco estados do Brasil

Resumo

Quarenta e cinco espécimes que representam nove espécies de répteis (*Salvator merianae*, *Enyalius bilineatus*, *Amphisbaena alba*, *Xenopholis undulatus*, *Chironius fuscus*, *Helicops angulatus*, *Chironius flavolineatus*, *Erythrolamprus viridis* e *Crotalus durissus*) coletados em cinco estados brasileiros foram examinados para helmintos. Foram encontrados doze espécies de helmintos sendo: nove Nematoda (*Physaloptera tupinambae*, *Strongyluris oscari*, *Paracapillaria* sp., *Dracunculus brasiliensis*, *Physaloptera liophis*, *Serpentirhabias* sp. 1, *Serpentirhabias* sp. 2, *Serpentirhabias* sp. 3 e *Aplectana* sp.), um Cestoda (*Semenoviella amphisbaenia*), um Trematoda (*Paracotyletrema* sp.) e um Acantocephala (*Centrorhynchus* sp.). Dez novos registros de hospedeiros e sete novos registros de localidade foram relatados.

Palavras-chave: parasita, serpentes, lagartos, anfisbenídeos, nematoda.

1. Introduction

Surveys is one of the most suitable ways to expand knowledge on biodiversity and understand how the species, and what species, are parts of an ecosystem. Likewise, parasitological surveys are important for understanding parasite/host relationships, as well as ecological aspects and estimations on local biodiversity (Rocha et al., 2003; Marcogliese, 2005; Campião et al., 2015). Moreover, parasitism is a key feature in population dynamics and community structure (Ernst and Ernst, 1980).

Inventories of helminths from reptiles are relatively frequent and are very useful for filling the gaps in the ecological and taxonomic knowledge, for they enhance and constitute the development and the comprehension of the parasitism in the vertebrates (Muzzall, 2005;

Ávila and Silva, 2010; Ávila et al., 2010; Pereira et al., 2012; Albuquerque et al., 2013; Cabrera-Guzmán and Garrido-Olvera, 2014).

Studies focusing helminth parasites infecting squamate reptiles in Brazil have been increased in the past few years (Bursey et al., 2007; Ávila et al., 2010; Ávila et al., 2011; Barreto-Lima et al., 2012; Araujo-Filho et al., 2014; Mati et al., 2015). The information gathered was also compiled on extensive literature reviews, such as Ávila and Silva (2010) for helminths of lizards and amphisbaenians and Fernandes and Kohn (2014) for trematodes of reptiles.

Herein, we report the helminths infecting nine species of reptile in seven localities from Brazil, thereby increasing our knowledge of reptile's parasites in the Neotropical region.

2. Materials and Methods

Reptiles were collected from 2002 to 2012 in five Brazilian states, Minas Gerais: Viçosa ($20^{\circ}45'16.52''S$; $42^{\circ}52'42.88''O$) and Florestal ($19^{\circ}53'18.68''S$; $44^{\circ}25'55.63''O$) municipalities, Mato Grosso: Aripuanã ($10^{\circ}10'35''S$; $59^{\circ}26'52.42''O$) municipality, Ceará: Várzea Alegre ($6^{\circ}47'39.68''S$; $39^{\circ}17'35.56''O$), Farias Brito ($6^{\circ}55'34.10''S$; $39^{\circ}34'23.83''O$), and Aiuba ($6^{\circ}33'54.38''S$; $40^{\circ}13'37.65''O$) municipalities, Pernambuco: Exú ($7^{\circ}26'14.56''S$; $39^{\circ}44'03.62''O$) municipality and São Paulo: Botucatu ($23^{\circ}3'17.56''S$; $48^{\circ}31'10.24''O$) municipality. Voucher specimens were housed in the Herpetological collections of the Universidade Federal de Minas Gerais (UFMG) and Museu de Zoologia da Universidade Federal de Viçosa (MZUFV), in Minas Gerais state, at the Coleção Zoológica de Vertebrados da Universidade Federal de Mato Grosso (UFMT) in Mato Grosso state, at the Coleção Zoológica da Universidade Regional do Cariri (URCA - P), in Ceará state.

Specimens were caught by hand or by pitfall-traps, euthanized, fixed in 10% formalin, and preserved in 70% ethanol. For each specimen, snout-vent length (SVL) was measured using a digital caliper (0.01 mm precision). The specimen o Chironius flavolineatus and Crotalus durissus were discarded due to stage of decomposition. Sex of the hosts was determined by direct inspection of gonads or presence/absence of hemipenes. The body cavity of all specimens other than those from Minas Gerais and São Paulo was opened by a longitudinal incision from throat to vent, the gastrointestinal tract was slit longitudinally, and the body cavity, lungs, gall bladder, lungs, stomach, and intestinal contents were removed and examined under a stereomicroscope. For specimens from Minas Gerais

state, parasites were collected during examination of gut content from three tegu lizards (*Salvator merianae*, Duméril and Bibron, 1839), and during formalin injection in one lizard (*Enyalius bilineatus*, Duméril and Bibron, 1837), one amphisbaenian (*Amphisbaena alba*, Linnaeus, 1758), and one snake (*Xenopholis undulatus*, Jensen, 1900). Helminths found in the gastrointestinal tract, lungs, or body cavity were placed in vials of 70% ethanol for later identification. For species identification, nematodes were cleared in phenol and acanthocephalans, trematodes, and cestodes were stained in carmine and cleared with creosote. All helminthes were analyzed using image analysis software (Qwin Lite 3.1, Leica Microsystems, Wetzlar, Germany). Voucher helminth specimens were deposited in the Coleção Helmintológica do Instituto de Biociências da Unesp de Botucatu (CHIBB) and Coleção Parasitológica da Universidade Regional do Cariri (URCA - P).

3. Results

A total of 45 specimens belonging to nine species of reptile were collected. Nineteen specimens were infected with at least one helminth species: *Salvator merianae* ($N = 3$, $SVL = 302.5 \pm 84.6$ mm), *Enyalius bilineatus* ($N = 1$, $SVL = 82$ mm), *Amphisbaena alba* ($N = 1$, $SVL = 480$ mm), *Xenopholis undulatus* ($N = 1$, $SVL = 3950$ mm), *Chironius fuscus*, Linnaeus, 1758 ($N = 1$, $SVL = 771$ mm), *Helicops angulatus*, Linnaeus, 1758 ($N = 16$, $SVL = 423 \pm 201$ mm), *Erythrolamprus viridis*, Günther, 1862 ($N = 10$, $SVL = 425 \pm 230$ mm), *Chironius flavolineatus*, Jan, 1873 ($N = 1$) *Crotalus durissus*, Laurenti, 1768 ($N = 1$). Two hundred and seventy two helminths belonging to 12 species were recovered (Table 1).

Table 1. Results containing the species, locality and number of species (n), Helminths, prevalence (P), distribution and Site of Infection (S.Inf.): Stomach (S), intestines (I), Large intestine (LI), Small intestines (SI), Buccal cavity (BC), Coelomic cavity (CC), Lung (L) and Body cavity (BoC).

Species	Locality (n)	Helminths	P	Distribution	S.Inf.
<i>Salvator merianae</i>	Viçosa-MG (3)	<i>Physaloptera tupinambae</i>	175	October 2002, October and November 2008	S/I
<i>Enyalius bilineatus</i>	Viçosa-MG (1)	<i>Strongyluris oscari</i>	16	November 2008	LI
<i>Amphisbaena alba</i>	Viçosa-MG (1)	<i>Semenoviella amphisbaenae</i>	26	February 2009	SI
<i>Chironius fuscus</i>	Aripuanã-MG (1)	<i>Paracotyletremia</i> sp.	1	October 2007	BC
<i>Chironius flavolineatus</i>	Botucatu-SP (1)	<i>Serpentirhabdias</i> sp. 1	3	May 2015	L
<i>Xenopholis undulatus</i>	Florestal-MG (1)	<i>Serpentirhabdias</i> sp. 2	13	October 2012	L
<i>Helicops angulatus</i>	Aripuanã-MG (3)	<i>Paracapillaria</i> sp.	5	October 2007	CC
		<i>Paracotyletremia</i> sp.	2	October 2007	BC
		<i>Dracunculus brasiliensis</i>	1	January 2008	CC
<i>Erythrolamprus viridis</i>	Várzea Alegre-CE (6) and Aiuba-CE (1)	<i>Centrorhynchus</i> sp.	10	October 2011 and May 2012	BoC
		<i>Physaloptera liophis</i>	8	October 2011, May and June 2012	S
		<i>Aplectana</i> sp.	2	October 2011 and May 2012	LI
<i>Crotalus durissus</i>	Botucatu-SP (1)	<i>Serpentirhabdias</i> sp. 3	10	May 2016	L

4. Discussion

Our study, 10 new host and 7 new locality records are provided. *Paracapillaria*, *Dracunculus* and *Physaloptera* are considered generalists, being frequently reported infecting amphibians and reptiles (Torres and Puga, 1996; Timi et al., 2007; Moravec and Santos, 2009; Pereira et al., 2012).

Nematodes of the genus *Paracapillaria* are known to use fishes as intermediate hosts (Gibbons, 2010), being reported infecting 39 fish species (De and Maity, 1994; Moravec et al., 1996; Timi et al., 2007), two amphibians (Goldberg and Bursey, 2002; Moravec et al., 2007), and 10 species of reptile (Vicente et al., 1993; De, 1998; Goldberg et al., 2004; Moravec and Gibson, 2007). *Helicops angulatus* and Mato Grosso state is a new host and new locality record for the genus *Paracapillaria*.

Dracunculus brasiliensis (Moravec and Santos, 2009) was described infecting the green anaconda *Eunectes murinus*, being the only species of *Dracunculus* reported in South American reptiles (Moravec and Santos, 2009). *Helicops angulatus* and Mato Grosso state is a new host and new locality record for the *D. brasiliensis*.

Physaloptera liophis (Vicente and Santos, 1974) was described in the stomach of *Liophis miliaris* from Brazil (Vicente and Santos, 1974) and latter recorded infecting *Bothrops neuwiedi* at the municipality of Ingá, Minas Gerais state, Brazil (Gouveia et al., 2012). *Erythrolamprus viridis* and Ceará state is a new host and new locality record for *P. liophis*. *Physaloptera tupinambae* (Pereira et al., 2012) can be distinguished from the other Neotropical species of *Physaloptera* by length of spicules and by possessing a bipartite internal tooth (Pereira et al., 2012).

Serpentirhabdias is specific of snakes (Tkach et al., 2014). The uncertain diagnostic value of various morphological characters has caused instability in the determination of species (Kuzmin et al., 2007; Tkach et al., 2014), but the presence or absence of various structures at the anterior end, as well as their variations in shape and position have been widely used for species differentiation this genus (Tkach et al., 2014). The distinctive shape of the frontal end, morphometric and morphological characters in this sample presented did not allow the comparative diagnosis with any of the described species of the genus. Some specimens found may represent new species; unfortunately, the poor condition or absence of males in the samples makes difficult their identification. This may be the case of *Paracotyletrema*, since only one species have been described from Uruguay (Volonterio et al., 2006). The genus is a new host record for *Chironius fuscus* and *Helicops angulatus* and new locality for Mato Grosso state.

Cistacanths of *Centrorhynchus* sp. have been reported infecting the intestine several amphibian and reptile hosts, they are paratenic host to bridge ecological gaps, because utilize arthropods as intermediate hosts, where larval development occurs. (Santos and Amato, 2010; Ávila and Silva, 2010). *Erythrolamprus viridis* and Ceará state is a new host and new locality record for the genus *Centrorhynchus*.

Aplectana is commonly found infecting amphibians and reptiles and actually comprises 51 species. Identification relies mainly on male characteristics such as spicule length and caudal papillae pattern (Sou and Nandi, 2015), and as we found only females the specific identity could not be assigned. *Erythrolamprus viridis* is a new host record for the genus *Aplectana*.

Semenoviella is a monotypic genus distinguished from *Oochoristica* by few characters, such as cirrus sac length and presence of vaginal sphincter (Rego, 1967). Minas Gerais state is a new locality record for *S. amphisbaenae* (Rudolphi, 1819).

In the past few years, many parasitological studies focusing Brazilian reptiles have been published (Araujo-Filho et al., 2014; Barreto-Lima and Anjos, 2014; Mati et al., 2015). However, 773 species of reptile species are currently recognized in Brazil (Costa and Bernils, 2015) and less than 30% were sampled for helminths. however, some species of snakes of parasites were only been described in recent years (Kuzmin et al., 2014, 2016) especially with inclusion of molecular data (Morais et al., 2016).

Thus, the data presented increases knowledge on the occurrence of species of parasites, their relations with the hosts, helping in knowledge related to the pattern of distribution of the species and here increase our knowledge about the helminth fauna associated with Brazilian reptiles.

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