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COMMUNITY STRUCTURE OF PARASITES OF THE TREE FROG *SCINAX FUSCOVARIUS* (ANURA, HYLIDAE) FROM CAMPO BELO DO SUL, SANTA CATARINA, BRAZIL

ESTRUCTURA DE LA COMUNIDAD PARASITARIA DE LA RANA ARBORICOLA SCINAX *FUSCOVARIUS* (ANURA, HYLIDAE) DE CAMPO BELO DO SUL, SANTA CATARINA, BRASIL

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ABSTRACT

Sixty specimens of *Scinax fuscovarius* (Lutz, 1925) were collected between May 2009 and October 2011 at Campo Belo do Sul, State of Santa Catarina, Brazil, and necropsied in search of helminth parasites. Only four helminth species were found: *Pseudoacanthocephalus* sp. Petrochenko, 1958, *Cosmocerca brasiliense* Travassos, 1925, *C. parva* Travassos, 1925 and *Physaloptera* sp. Rudolphi, 1819 (larvae). The genus of the female cosmocercids could not be determined. Only 30% of the anurans were parasitized. *Scinax fuscovarius* presented low prevalence, infection intensity, and parasite richness. Sex and size of *S. fuscovarius* individuals did not influence the prevalence, abundance, and species richness of helminth parasites.

Keywords: Acanthocephala - anurans - helminth parasites - hylids - Nematoda - tree frogs

RESUMEN

Sesenta ejemplares de *Scinax fuscovarius* (Lutz, 1925) fueron colectados entre mayo de 2009 y octubre de 2011 en Campo Belo do Sul, Estado de Santa Catarina, Brasil y necropsiados para el estudio de sus helmintos parásitos. Fueron halladas solamente cuatro especies de helmintos: *Pseudoacanthocephalus* sp. Petrochenko, 1958, *Cosmocerca brasiliense* Travassos, 1925, *C. parva* Travassos, 1925 and *Physaloptera* sp. Rudolphi, 1819. Las hembras de cosmocércidos no pudieron ser determinadas debido a la falta de caracteres taxonómicos. Solamente el 30% de los anuros estuvieron parasitados. *Scinax fuscovarius* presentó baja prevalencia, intensidad de infección y riqueza parasitaria. El sexo y el tamaño de *S. fuscovarius* no influyeron en la prevalencia, abundancia y riqueza de especies de helmintos parásitos.

Palabras clave: Acanthocephala – anuros - helmintos parásitos – hílidos – Nematoda – rana arborícola

INTRODUCTION

There are a few factors, such as host habit and diet that may influence parasite community structure. Anurans are a much diversified group of vertebrates with 7300 extant species which can be fossorial, arboreal, terrestrial, semi-aquatic or aquatic (Duellman & Trueb, 1994; Frost, 2014). Aho (1990) observed that semi-aquatic anurans had a tendency to present greater helminth richness; terrestrial and aquatic anurans presented intermediate and similar values, while fossorial and arboreal anurans presented lower helminth richness.

There are some studies on anurans that demonstrate a positive relationship between the host habitat and the variability of the parasite community structure (Hamann & Kehr, 1998; Bolek & Coggins, 2000; Muzzal *et al.*, 2001; Bursey *et al.*, 2001; Iannacone, 2003; León-Règagnon *et al.*, 2005; Luque *et al*., 2005; Brooks *et al.*, 2006; Goldberg *et al*., 2007; González & Hamann, 2008; González, 2009; Santos & Amato, 2010; Hamann *et al.*, 2010; Campião *et al.*, 2010; Santos *et al.*, 2013). Aquatic and/or semi-aquatic anurans tend to present a greater number of digenean species, because most of the anuran digeneans use aquatic or semi-aquatic arthropods as secondary intermediate hosts (Prudhoe & Bray, 1982).

The diversity of parasite communities of the Neotropical anurans is unknown. Since this region concentrates the largest number of global anuran richness, an adequate description of their parasite diversity is important to better understand some patterns that are not well defined. Luque *et al.* (2005), Santos & Amato (2010), Santos *et al.* (2013) and Chero *et al.* (2014) presented results of recent studies on *Rhinella icterica* (Spix, 1824), *Rhinella fernandezae* Gallardo, 1957 and *Telmatobius jelskii* (Peters, 1863) demonstrating the contribution of this kind of research and showing that the parasite richness is higher than that cited by Aho (1990).

Scinax fuscovarius (Lutz, 1925) occurs from south to southern Brazil and eastern Argentina, Paraguay and Bolivia. It is found in open areas such as pastures. At night during the reproduction season they are found in still water (lakes, ponds and wetlands), while at other periods they take refuge in trees. The tadpoles are nektonic and feed on suspended matter. As adults, males measure from 37 to 47 mm, and females from 42 to 48 mm (Kwet & Di-Bernardo, 1999; Kwet *et al.*, 2010). This species is very abundant in several areas of southern Brazil, nevertheless, little is known about its parasite community structure. Being an arboreal species, *S. fuscovarius* is a good model to analyze if arboreal frogs have lower helminth richness as proposed by Aho (1990).

This study aimed to: identify the helminth parasites of *S. fuscovarius*; analyze the parasite community structure; and increase the knowledge of the biology of these species with parasitological data.

MATERIAL AND METHODS

Sixty *S. fuscovarius* specimens (28 females, 31 males, one specimen with undetermined sex), were captured under license SISBIO (Number 026/2006), between June 2009 and December 2011, at the Municipality of Campo Belo do Sul (27°59' 42.19"S, 50°53'27.92"W, ranges between 700-900 m.a.s.l), located in the Araucarias Plateau Region of the State of Santa Catarina, southern Brazil.

The specimens were manually collected and sacrificed with lidocaine Geyer® 2% (local anesthetic), spread on the abdomen of the animal and absorbed by the skin; they were weighed and measured (snout-cloacal length). The sex of the host was identified through the

coloration of the vocal sac (dark in males) and gonad examination of juveniles.

Helminths were collected and placed in physiological saline solution 0.65%; they were fixed, stained and/or diaphanized following Amato & Amato (2010). The morphologic and biometric study was performed using an Axiolab Zeiss microscope. The ecological descriptors, prevalence, infection intensity, parasite abundance and richness followed Bush *et al.* (1997).

Statistical analysis included only the parasite species with prevalence greater than 10% (Bush *et al.*, 1997). The Mann-Whitney *"U"* test was calculated to evaluate if the sex of the host was influenced by parasite abundance and richness. Fisher's exact test was used to evaluate if the host's sex and size influenced parasite prevalence. To evaluate if host size could influence the prevalence of helminth species, anurans were categorized as small or large frogs.

The Spearman correlation coefficient (r_s) evaluated the influence of size on the parasite abundance and richness of the analyzed helminth hosts. The Berger-Parker index (*d*) was used with a numerical measure of dominance (Nering & Von Zuden, 2010). The representativity of helminth species richness found was analyzed with the program EstimateS 8.0 (Colwell, 2009) through the collectors curve and the *Chao 1* Index and *ACE* ("Abundance-based Coverage Estimator"), estimators based on abundance that quantify rarity, and estimators based on incidence, *ICE* ("Incidence Coverage Estimator-based") and *Jackknife*. Female Cosmocercidae were not considered in the species richness representativity analysis since these specimens did not show diagnostic characters that allow separation between *Cosmocerca* and *Aplectana*.

Representative helminth specimens were vouchered at the Helminthological Collection of Instituto Oswaldo Cruz (CHIOC), FIOCRUZ, Rio de Janeiro, Rio de Janeiro State, Brazil. All hosts examined were vouchered at the Amphibian Collection, Herpetology Laboratory, Zoology Department, Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil.

RESULTS

A total of 35 helminths, representing an average intensity of infection of 0.6 helminths / host, were collected. Only acanthocephalans and nematodes were found (Table 1). The parasite community was composed of four helminth species, *Pseudoacanthocephalus* sp., *Cosmocerca parva* Travassos, 1925 and *Cosmocerca brasiliense* Travassos, 1925, and one species was represented by only a larval form, *Physaloptera* sp. Due to the lack of taxonomic characters the female Cosmocercidae Railliet, 1916 could not be determined to genus and species. Acanthocephalans were not determined to the specific level because no males were found. Besides the characteristics of the proboscis, it is necessary to observe the morphology and the number of cement glands of *Pseudoacanthocephalus* species.

Of the 60 anurans collected, only 18 were infected with parasite species. Nematodes accounted for 80% of the sample (three species), and 20% were represented by one acanthocephalan species. A total of 23 nematodes and 12 acanthocephalans were collected. The nematodes infected 15 hosts while acanthocephalans infected only three hosts.

Only Cosmocercidae females showed prevalence greater than 10% and were the dominant parasites (d = 0.47) of the sample. There was no influence of the sex of the host on

the prevalence (p = 0.30), abundance (*"U"* = 372; p = 0.34) and parasite richness (*"U"* = 436; p = 0.85), and the host size did not influence the prevalence (p = 0.49), abundance (r_s = 0.14; p = 0.27) and parasite richness (r_s = 0.17; p = 0.19).

The collector curve (SOBs) did not reach the asymptote; on the other hand, the estimators used *ACE*, *ICE*, *Chao 1* and *Jacknife* reached stability (Figure 1), which indicates that 80% to 100% of helminth species were sampled in this study.

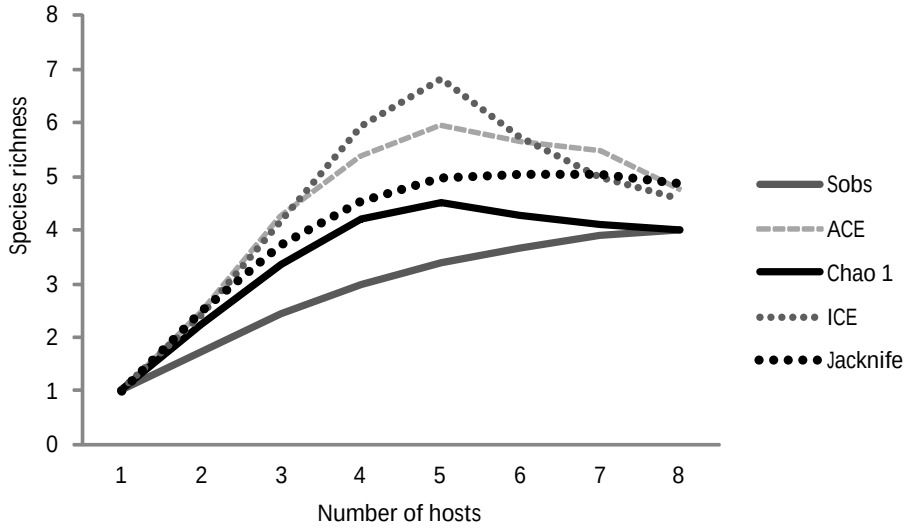

Figure 1. Accumulated richness observed (SOBs) and estimated (*ACE*, *Chao 1*, *ICE* and *Jacknife*) of the helminth community of *Scinax fuscovarius*, Campo Belo do Sul, State of Santa Catarina, Brazil.

Table 1. Prevalence, intensity, mean intensity, mean abundance and infection site of parasite helminths of *Scinax fuscovarius* (n = 60), Campo Belo do Sul, State of Santa Catarina, Brazil.

Parasites	Prevalence (%)	Intensity	Mean intensity ±SD	Mean abundance ±SD	Infection site
Acanthocephala					
Pseudoacanthocephalus sp. CHIOC 38283; 38284	5	1 to 8	4,00 ± 3,61	0,2 ± 1,10	Small intestine
Nematoda					
Cosmocercidae CHIOC 38157	16,6	1 to 2	1,80 ± 1,87	0,3 ± 1,00	Small/large intestine
Cosmocerca parva CHIOC 38158	2	1	1	0,03 ± 0,18	Small/large intestine
Cosmocerca brasiliense CHIOC 38159	2	1	1	0,03 ± 0,18	Small/large intestine
Physaloptera sp. (larvae) CHIOC 38160	1,66	1	1	0,02 ± 0,13	Stomach

CHIOC = Helminth Collection of Instituto Oswaldo Cruz, with the numbers of representative specimens.

Table 2. Parasite richness of anurans collected in the municipality of Campo Belo do Sul, State of Santa Catarina, Brazil.

Rhinella icterica Terrestrial	*Leptodactylus latrans* Semi-aquatic	*Physalaemus cuvieri* Terrestrial	*Scinax fuscovarius* Arboreal
Digenea	**Digenea**	**Digenea**	**Acanthocephala**
Catadiscus cohni	*Catadiscus cohni*	Metacercária (larva)	*Pseudoacanthocephalus* sp.
Rudolphitrema rudolphi	*Catadiscus pygmaeus*	**Eucestoda**	**Nematoda**
Eucestoda	*Catdiscus inopinatus*	*Cylindrotaenia americana*	Cosmocercidae
Cylindrotaenia americana	*Catadiscus uruguayensis*	**Monogenea**	*Cosmocerca parva*
Plerocercóide (larva)	*Gorgoderina parvicava*	*Polystoma cuvieri*	*Cosmocerca brasiliense*
Acanthocephala	*Gorgoderina megacysta*	**Nematoda**	*Physaloptera* sp. (larva)
Cistacanto (larva)	*Choledoscystus elegans*	*Strongyloides* sp.	
Nematoda	*Rauschiella linguatula*	Cosmocercidae	
Strongyloides sp.	**Eucestoda**	*Cosmocerca rara*	
Rhabdias fuelleborni	*Cylindrotaenia americana*	*Cosmocerca parva*	
Cosmocercidae	Plerocercóide	*Cosmocerca cruzi*	
Cosmocerca rara	**Nematoda**	*Aplectana elenae*	
Cosmocerca brasiliense	*Strongyloides* sp.		
Aplectana elenae	*Rhabdias elegans*		
Oxyascaris oxyascaris	Cosmocercidae		
Nematoide não identificado	*Cosmocerca rara*		
	Cosmocerca parva		
	Cosmocerca cruzi		
	Cosmocerca brasiliense		
	Aplectana elenae		
	Oxyascaris oxyascaris		

DISCUSSION

Analysis showed that *S. fuscovarius* had low parasite richness, which is probably related to the species' habitat. The nematodes found showed low prevalence and low infection intensities, and most hosts were parasitized with only one helminth species.

The parasite richness of *S. fuscovarius* is different from those found in *Rhinella icterica* (Spix, 1824) (Santos *et al.*, 2013), *Leptodactylus latrans* (Steffen, 1815) and *Physalaemus cuvieri* Fitzinger, 1826, collected on the same area (Table 2); all four species have distinct habits. However, the result obtained in this study corroborates Hamann & Kehr (1998), Bursey *et al.* (2001), Goldberg *et al.* (2002) and González & Hamann (2008), which also found low values of parasite richness on hylid anurans of South America.

The nematode species *C. parva* and *C. brasiliense* found in this study have direct cycles (monoxenous) and infect their hosts through cutaneous penetration. Species of *Physaloptera* have an indirect cycle (heteroxenous) and are commonly found in anurans which, according to Anderson (2000), act as paratenic hosts for *Physaloptera* species, while lizards, snakes and mammals act as definitive hosts.

According to Bursey *et al.* (2001), *C. brasiliense* is the most common and greater prevalence parasite species found on arboreal anurans. Goldberg *et al*. (2007) working in the State of Pará, Brazil found low prevalence (10%) and an intensity equal to 1 for the parasite *C. brasiliense* in the species *Dendropsophus cachimbo* (Napoli and Caramashi, 1999). González & Hamann (2008) found *C. parva* and *Physaloptera* sp. (larval form) in *Scinax acuminatus* at Corrientes, Argentina, which also showed low prevalence.

In this study, the three parasite species (*C. brasiliense*, *C. parva* and *Physaloptera* sp.) showed low prevalence and infection intensity. On hylids the low vagility and the arboreal habit probably contributes to a low infection occurrence by nematodes with cutaneous penetration (Hamann *et al.*, 2010).

The genus *Pseudoacanthocephalus* Petrochenko,1956 includes only parasite species of amphibians and reptiles. In South America, there are records of *Pseudoacanthocephalus lutzi* (Hamann, 1891) (= *Acanthocephalus lutzi* (Hamann, 1891); = *Acanthocephalus saopaulensis* Smales, 2007) in Brazil (Hartwich, 1956; Pinhão *et al.*, 2009), Argentina (Lajmanovich & Ferrato, 1995), Peru (Tantaléan, 1976), Uruguay (Cordero, 1933; Hartwich, 1956) and Paraguay (Smales, 2007). The infection by this acanthocephalan occurs by the ingestion of an arthropod; Arredondo & Pertierra (2009) suggest that aquatic insects are the intermediate hosts of *P. lutzi*.

Hamann *et al.* (2010) analyzed the parasite community structure of *Scinax nasicus* from Corrientes, Argentina, and found that this host had high richness of helminth parasite species, with 21 species. However, only seven species were adult forms while the others were larval forms. The results observed by these authors demonstrate the important role that this anuran species plays in the food chain, occupying an intermediate position between the parasite and its definitive hosts (snakes, birds and mammals).

Usually, the sex of the anuran does not influence the structure of the parasite community, once they are evenly distributed between males and females. This pattern was found on *S. fuscovarius* in which sex did not influence the parasite richness, the helminth prevalence and abundance. This result

corroborates other studies (Yoder & Coggins, 1996; McAlpine, 1997; Barton, 1999; Goldberg *et al.*, 2002; Hamann *et al.*, 2006a,b; Santos & Amato, 2010; Santos *et al.*, 2013).

On the other hand, some authors suggest that the host size influences the parasite richness and the helminth prevalence and abundance (Baker, 1984; McAlpine, 1997; Gillilland & Muzzall, 1999; Bolek & Coggins, 2003; Chero *et al.*, 2016). In this study, however, this correlation was not observed, since size did not influence the parasite community structure of *S. fuscovarius*, this result corroborates with Chero *et al.* (2014). This result may indicate that, regardless of size, all individuals are susceptible to infection by Cosmocercidae.

The estimators did not reach an asymptote and this result may be related to the low number of infected anurans, but the estimated values did not extrapolate and remained close to observed. The parasite richness of *S. fuscovarius* is, probably, between four and five species.

The results obtained in this study was corroborated by Hamann *et al.* (2010) who suggested that the sit-and-wait strategy and the particular diet of these anurans are determining factors for the low parasite richness found. The limited movement and the arboreal habit of this species also contribute to the low occurrence of nematodes that parasitize anurans through cutaneous penetration. The absence of digenetic is probably related to the little contact of these anurans with the aquatic environment.

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