Helminth community dynamics in a population of *Pseudopaludicola pocoto* (Leptodactylidae: Leiuperinae) from Northeast-Brazilian

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Summary

Climatic variation in low latitudes influences the dynamics and structure of parasite communities. Environmental changes caused by dry and rainy seasons alter prevalence and abundance of endoparasite communities. In addition to providing a list of the helminth species associated with the swamp frog *Pseudopaludicola pocoto*, this study aimed to investigate the effects of rainfall and temperature on parasitological descriptors of helminths associated with *P. pocoto* in an area of the semiarid zone. A total of 817 swamp frog specimens were collected between 2013 and 2017, with four sampling expeditions during the dry season and four during the rainy season. Environmental parameters of temperature and rainfall were compared to the parasitological descriptors of prevalence, abundance and mean infection intensity of the parasite community using a multivariate linear regression. A richness of eight parasite species was identified, including Nematoda (*Rhabdias* sp., *Cosmocerca parva*, *Oxyascaris oxyascaris*, *Physaloptera* sp., *Brevimulticaecum* sp*., Spiroxys* sp. and unidentified nematode) and Acanthocephala (cystacanths). Rainfall levels had a significant effect on the infection intensity of *Rhabdias* sp. being the presence of this species higher during the rainy season, whereas no influence of temperature was observed on the helminth community.
Keywords: Anura; Caatinga; helminthfauna; Neotropical; seasonality; semiarid

Introduction

Leptodactylidae is one of the ubiquitous frog families in the Neotropics, with great richness and abundance in the Caatinga biome (Roberto *et al*., 2013; Ávila, 2015). Leptodactylids occur in a wide variety of habitats, becoming exposed to several degrees of helminth infections (Goldberg *et al*., 2007; Bursey & Brooks, 2010; Hamann & González, 2010). The genus *Pseudopaludicola* currently comprises 22 species of small swamp frogs distributed in South America (Cardozo *et al*., 2018). To date, only two species

had their helminth fauna investigated – *Pseudopaludicola boliviana* Parker, 1927, in which a richness of ten taxa was found, including trematodes, cestodes, nematodes and acanthocephalans (Duré *et al*., 2004; González & Hamann, 2012), and *Pseudopaludicola falcipes* Hensel, 1867, in which only the nematode *Cosmocerca podicipinus* Baker and Vaucher, 1984 was recorded (González & Hamann, 2004; 2009). *Pseudopaludicola pocoto* Magalhães, Loebmann, Kokubum, Haddad & Garda, 2014 was recently described from Northeast-Brazilian and is widely distributed in the Caatinga biome, however, there are still no studies on its ecology, only on

its geographical distribution (Magalhães *et al*., 2014; Pereira *et al*., 2015; Silva *et al*., 2015; Lantyer-Silva *et al*., 2016 and Silva *et al*., 2017).

Several factors contribute to the dynamics and structure of parasite communities, like seasonality, environmental heterogeneity or factors associated with the host, such as spatial distribution, population density, and body size (Aho, 1990). Climate changes can cause some effects upon biological communities, like alterations on the abundance and transmission rates of helminths and also have an influence on host-parasite relations (Altizer *et al*., 2006; Koprivnikar *et al*., 2006; King *et al*., 2007; Koprivnikar & Poulin, 2009; Schotthoefer *et al*., 2011; Pizzato *et al*., 2013 and Brito *et al*., 2014). The prevalence and abundance of helminths are more influenced by seasonal variations in regions of median latitudes because cold seasons alter the acquisition of the parasite by the host (Pizzato *et al*., 2013). Meanwhile, in tropical areas, the prevalence and abundance of parasites can either increase or decrease between dry and rainy season (Choudhury & Dick, 2000). Thus, climatic factors, such as temperature, humidity, and rainfall levels can exert different effects upon parasitological descriptors of helminth infections.

Identifying what are the environmental factors that influence the helminth community can contribute towards the comprehension of how infection dynamics changes through time and space in order to unravel the mechanisms involved in host-parasite interactions. Besides providing a list of the helminths associated with *P. pocoto*, this study aims (I) to compare the similarity between the helminth communities associated with species of *Pseudopaludicola* and (II) to investigate the effects of rainfall and temperature upon the parasitological descriptors of prevalence (P), mean intensity of infection (MI), abundance, diversity, and migration of parasites between sites of infection in the helminths community associated with *P. pocoto* in Brazilian Northeast.

Material and Methods

This study was carried out in the Benguê Reservoir, Aiuaba, Ceará, Brazil (06°35'35"S, 40°08'31"W). Host samplings were performed from September 2013 to March 2017, with four expeditions during the dry season and four during the rainy season. This region is located in one of the driest areas of Ceará State, with mean annual rainfall levels of 560 mm (Funceme, 2016).
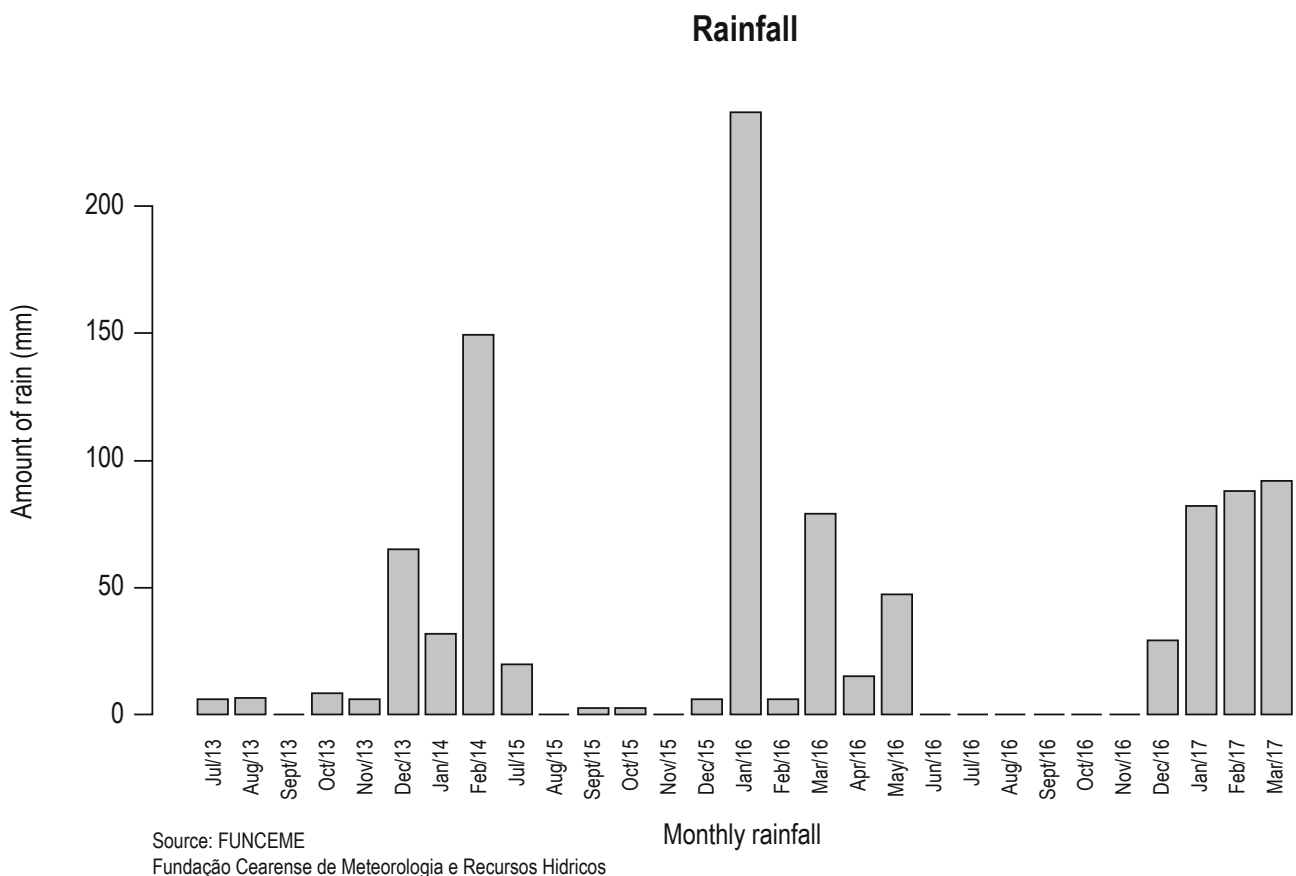
Rainfall

Source: FUNCEME
Fundação Cearense de Meteorologia e Recursos Hidricos

Fig. 1. Monthly rainfall levels related to the sample period of *P. pocoto* in the municipality of Aiuaba, Ceará State, Brazil.
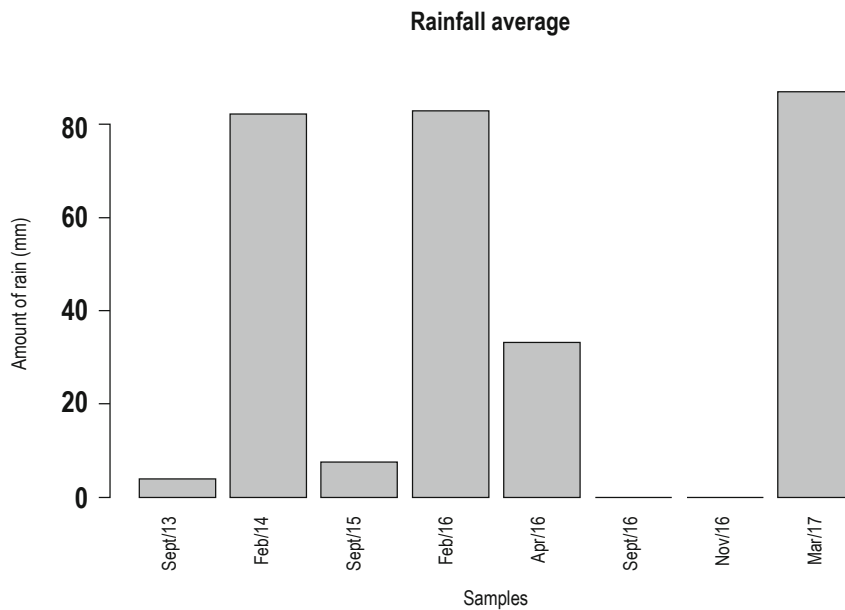
Rainfall average

Fig. 2. Quarterly means of rains related to the sampling months of *P. pocoto* in the municipality of Aiuaba, Ceará State, Brazil.

A total of 817 specimens of *P. pocoto* (573 males, mean snout-vent length [SVL] ± SD 13.97 ± 1.56 mm, range: 10.15 – 16.5 mm, 244 females, SVL: 15 , 1 ± 1,57 mm, range: 11,41 – 18,46 mm) were used for this study, being collected by hand for this parasitological and also specimens collected for other purposes and deposited in the Herpetology Collection of the Universidade Regional do Cariri - URCA-H, Crato, Ceará State were used. Specimens were euthanized with sodium thiopental, necropsied for helminths, fixed in 10 % formaldehyde and stored in 70 % ethanol.

Data on rainfall levels were gathered using monthly means from the Fundação Cearense de Meteorologia e Recursos Hídricos – FUNCEME (Foundation of Meteorology and Hydric Resources of Ceará State). For statistic analyses between rainfall and parasitological descriptors, a mean rainfall of every three months was extracted related to the period between the samplings (Figs. 1 and 2).

The following parasitological descriptors: prevalence, mean intensity of infection, and abundance were calculated following Bush *et al*. 1997 using standard error and range. Aggregation of parasites was calculated using the Discrepancy Index (D) by Poulin 1998 which ranges from 0 to 1, where D = 0, all hosts harboring the same number of parasites; D = 1, all parasites found in a single host. This index was calculated in the software Quantitative Parasitology 3.0 (Rózsa *et al*., 2000).

Nematodes were found alive, washed in saline solution (0.9 % NaCl), fixed and preserved in 70 % ethanol. The nematodes were cleared in lactophenol or lactic acid while acanthocephalans were removed from their cysts, stained in carmine, and cleared in creosote. All endoparasites were observed and identified to the lowest possible level under a light microscope DMLB (Leica) and DM 5000B with interferential phase contrast, according to the litera-

Table 1. Helminth component community associated with *Pseudopaludicola pocoto* from the municipality of Aiuaba, Ceará State, Brazil.

Helminth	P (%)	MI	MA	IS	Stage	Range
Nematoda						
Rhabdias sp.[a]	22.6	1.49 ± 1.7	0.34	Lungs	Adult	1 – 8
Cosmocerca parva [a,b]	25.5	1.35 ± 1.4	0.34	St, SI and LI	Adult	1 – 5
Oxyascaris oxyascaris [a,b]	12.3	1.54 ± 1.8	0.2	SI and LI	Adult	1 – 7
Physaloptera sp.[a]	0.1	1 ± 1	0	St	Larva	1
Spiroxys sp.[a]	0.1	1 ± 1	0	Cav	Larva	1
Brevimulticaecum sp.[a,b]	0.1	1 ± 1	0	Cav	Larva	1
Unidentified nematode	2.9	1.5 ± 2.3	0.4	St, SI and Cav	Larva	1 – 4
Acanthocephala						
Cystacanth	1.1	1.43 ± 1.79	0	Cav	Cyst	1 – 3

P (%) - prevalence; MI – mean intensity of infection; MA – mean abundance; IS – infection site; St – stomach; SI – small intestine; LI – large intestine; Cav – body cavity; a – new record; b – new locality

ture (Yamaguti, 1961; Sprent, 1979; Vicente *et al.*, 1991; Anderson, 2000 and Gibbons, 2010). All parasites were deposited at the Coleção Parasitológica do Laboratório de Zoologia from Universidade Regional do Cariri – LZ-URCA (Parasitological Collection of the Laboratory of Zoology).

Richness (total number of helminths) and Brillouin's index of diversity were used to describe the parasite community. Richness was estimated using species accumulation curve, in which the number of observed species is a function of the sampling effort, measured in number of individuals using the R software packages Biodiversity R and Vegan (R core team, 2014). The Shapiro-Wilks test was applied to evaluate the normality of prevalence data, mean intensity of infection, mean abundance (MA) and diversity. Thereby, diversity between seasons was compared using *Wilcoxon's* test for paired samples and differences of prevalence, mean intensity of infection, and mean abundance between seasons was tested by Student *t*-test.

Using a data matrix with the presence/absence variables for the parasite species related to the genus *Pseudopaludicola*, the degree of similarity among these helminth communities was calculated using the Sorensen's index (So), with a posterior analysis of clustering using the Cluster method using the unweighted pairgroup average (UPGMA).

A multivariate regression was performed to assess the influence of rainfall levels and temperature and their interaction on prevalence, mean intensity of infection and abundance of the helminth community of *P. pocoto*. To evaluate whether there were alterations of infection sites between dry and rainy seasons, a contingency table was made with data on the abundance of species infection/sites. The helminths that did not vary sites between the seasons and the ones that were not frequent were excluded from the analyses not to have influence of these values upon the species that showed greater abundance and occupied different sites. To evaluate the significance of the results, a chi-square test was performed with the data organized as a contingency table of two factors (Gotelli & Ellison, 2011). All statistical analyses were performed using the software PAST 3.0.

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed according collection authorization issued by Chico Mendes' Institute (ICMBio/SISBio) Nº 29613 – 1; 55467 – 1 for scientific activities aims and authorized by council from Universidade Regional do Cariri-Urca nº 00260/2016.1.

Results

From the 817 hosts necropsied, 406 were parasitized with at least on helminth taxon (P = 49.7 %, MI = 1 ± 0.51, MA = 0.49 ± 0.4, range = 1 – 8). From the 405 hosts sampled in the dry season, 193 were parasitized with at least one helminth taxon (P = 47.7 %,
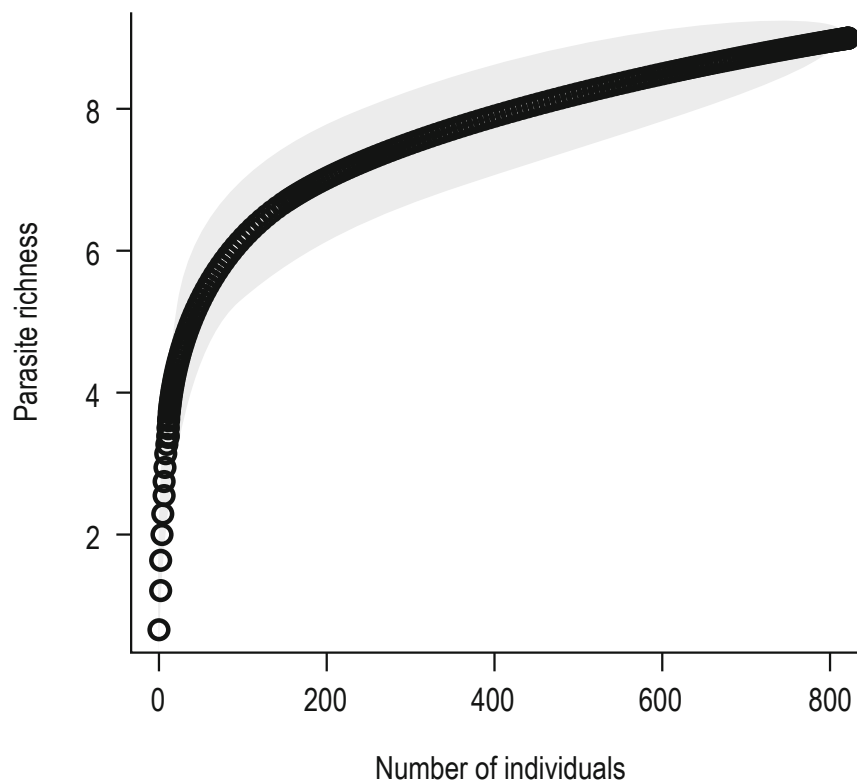

Fig. 3. Species accumulation curve (black line) and confidence interval (gray) for the richness of the helminths associated with *P. pocoto*, from the municipality of Auiaba, Ceará State, Brazil.

Table 2. Effects of rainfall levels and temperature on the infection descriptors of the helminth community of *Pseudopaludicola pocoto*, from the municipality of Aiuaba, Ceará State, Brazil.

		Prevalence	Infection Intensity	Abundance
Rainfall				
Helminth community	r	0.195	0.632	0.427
	p	0.64	0.09	0.29
Rhabdias sp.	r	0.464	0.91	0.556
	p	0.24	0.0014*	0.15
C. parva	r	0.118	0.574	0.284
	p	0.78	0.14	0.49
O. oxyascaris	r	0.298	0.02	0.372
	p	0.47	0.95	0.36
Temperature				
Helminth community	r	-0.428	0.672	-0.462
	p	0.29	0.07	0.25
Rhabdias sp.	r	-0.582	-0.41	-0.5
	p	0.13	0.31	0.2
C. parva	r	-0.485	-0.611	-0.54
	p	0.22	0.11	0.17
O. oxyascaris	r	-0.265	0.371	-0.31
	p	0.52	0.36	0.45
Rainfall: Temperature				
Helminth community	r	0.03	0.4	0.18
	p	0.59	0.14	0.48

*Effects of rainfall levels on infection intensity of *Rhabdias* sp. (p-value < 0.05)

MI = 1 ± 0.52, MA = 0.48 ± 0.6, range = 1 – 5), and from the 412 hosts sampled in the rainy season, 213 were parasitized with at least one helminth taxon (P = 51.7 %, MI = 1 ± 0.48, MA = 0.52 ± 0.6, range 1 – 8).

A total of 803 helminths specimens were collected, including nematodes and acanthocephalans, showing a richness of eight taxa (*Rhabdias* sp., *Cosmocerca parva* Travassos, 1925, *Oxyascaris oxyascaris* Travassos, 1920, *Physaloptera* sp., *Brevimulticaecum* sp., *Spiroxys* sp., unidentified nematode, and cystacanths) (Table 1). The infracommunity richness varied from one to three helminth species by host. The most abundant species recorded in this study were found in adult stage: *Rhabdias* sp. (N = 279), *C. parva* (N = 285), and *O. oxyascaris* (N = 157).

The aggregation index of the helminths showed moderate values in the dry season (*D* = 0.52±0.5) and rainy season (*D* = 0.48±0.48). The Brillouin's diversity index for the dry and rainy season were *i* = 1.42 and *i* = 1.29, respectively. There was no difference between seasons for prevalence (t= -0.0439; p = 0.96), mean intensity of infection (t = 1.359; p = 0.27) and mean abundance (t = -0.824; p = 0.47). The accumulation curve and the confidence interval showed a tendency to stabilization of richness of the helminths associated with this host in that location (Fig. 3).

The similarity between the communities of helminths of *P. pocoto* and *P. boliviana* and between *P. pocoto* and *P. falcipes* was of (So = 0), totally differing in their compositions of parasitic species. The proximity between *P. boliviana* and *P. falcipes* was of (So = 18.1 %). The distance between the communities analyzed and compared with the present study are represented in Figure 4.

There was no influence of rainfall or temperature or the interaction between both environmental variables on the community of
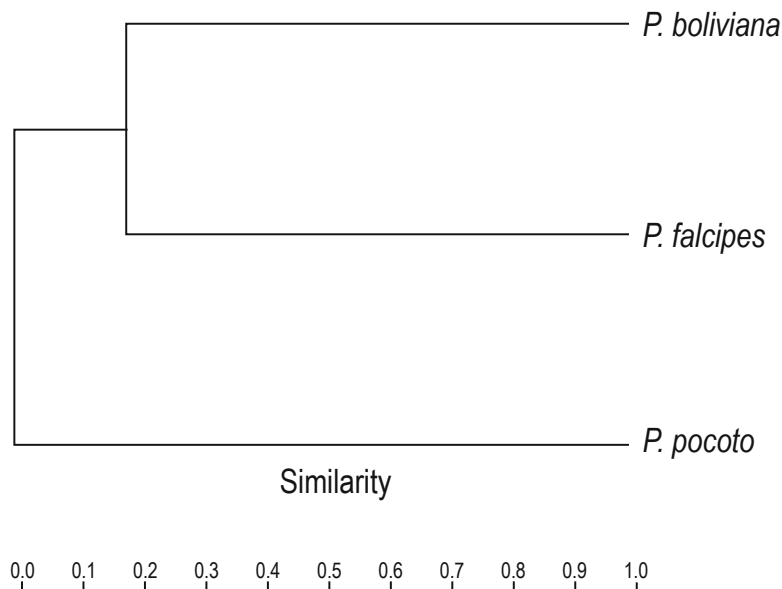

Fig. 4. Dendogram based on Sorensen similarity index comparing the helminth communities associated by genus *Pseudopaludicola*.
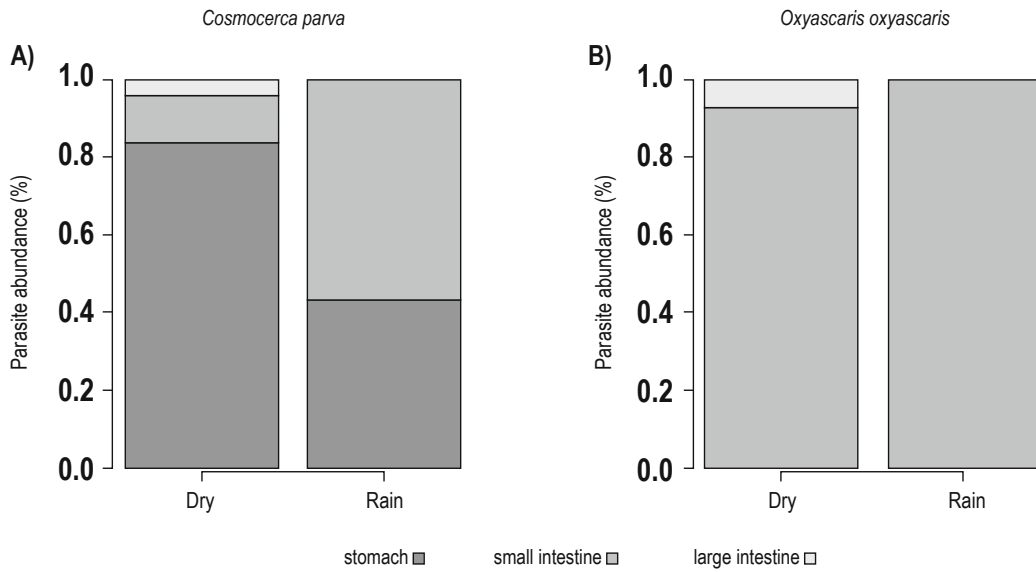
Fig. 5. Species/site relation between dry and rain season.

helminths *P. pocoto* (Table 2). However, the rainy season had a significant influence on intensity of infection of *Rhabdias* sp. (p = 0.0014), which was not found for the other abundant species (Table 2).

Regarding range of infection sites, Figure 5 shows the results of the most abundant species and the ones with greatest variation of sites. The chi-square test showed significant differences of the parasites migration patterns among the infection sites of the hosts between dry and rainy season for *C. parva* (p = 9.84e^{-17}). *Cosmocerca parva* was more related to the stomach (n=129) in the dry season, while in the rainy season this species used both

stomach (n=67) and the intestines (n=89). However, for *O. oxyas-caris* (p = 0.0024), although significant, the test found variation in the abundance of infection of this species between dry and rainy season, but no changes in the infecting site. The values of abundance in each host's site infection by season and total values are presented in Figure 6.

Discussion

Similar to other members of the Leptodactylidae, *P. pocoto* prefers semi-aquatic habitats (Frost, 2013), which is mirrored in the
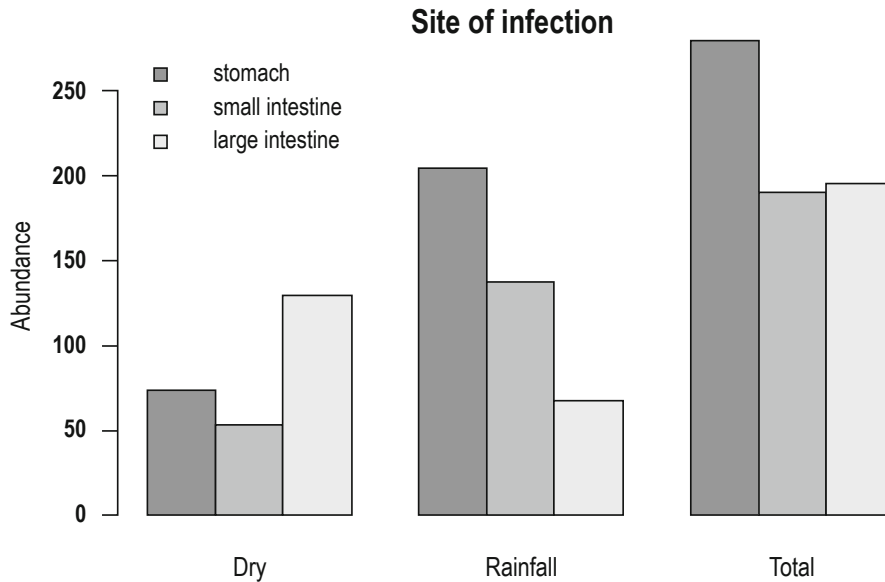
Site of infection

Fig. 6. Abundance values of infection by host site.

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Table 3. A selection of parameters in male individuals of *Cosmocerca parva*.

	Cosmocerca parva	
	Travassos, 1925	Present study
Body length	1.42 – 2.01	1.38
Spicule length	90 – 110	89
Gubernaculum length	85 – 108	69
Number of plectanes	5 – 7	5
Lateral alae	Present	Present

Column 1 represents the metric range in μm of the morphometry of *C. parva*. (Revised from Bursey et al., 2015)

infection routes of its helminths, since the most abundant parasite species found in this study have direct life cycle. Nematodes were the most frequent taxa in the helminth community of *P. pocoto* in this study, with representatives of six families (Rhabdiasidae, Cosmocercidae, Oxyascarididae, Physalopteridae, Gnathostomatidae, Heterocheilidae). As in the present study, cosmocercids are the most frequently nematodes found among Leptodactylidae (Duré *et al.*, 2004; González & Hamann, 2004; 2012; Santos & Amato, 2013; Campião *et al.*, 2014).

The genus *Rhabdias* currently includes approximately eighty species, of which fifteen are valid for the Neotropical region (Kuzmin *et al.*, 2016). From the species that occur in Brazil, seven are found in the amphibians and reptiles: *Rhabdias androgyna* Kloss, 1971, *Rhabdias fuelleborni* Travassos, 1926, *Rhabdias hermafrodita* Kloss, 1971, *Rhabdias galactonoti* Kuzmin, Melo, Silva-Filho and Santos, 2016, *Rhabdias paraenses* Santos, Melo, Nascimento, Nascimento, Giese and Furtado, 2011, *Rhabdias breviensis* Nascimento, Gonçalves, Melo, Giese, Furtado and Santos, 2013, and *Rhabdias stenocephala* Kuzmin, Melo, Silva-Filho and Santos, 2016 (Kuzmin *et al.*, 2016).

The morphological and morphometric characters often used for the characterization of *Rhabdias* species largely overlap (Tkach *et al.*, 2014). Currently, the most promising morphological characters with a tendency to accompany molecular data results are the shape and structure of the apical region, which are classified into five categories: (a) absence of lips, (b) six lips uniform in size and shape (c) four submedian and two pseudolabial lips, (d) two lateral pseudolabia and four in protuberance forms, and (e) species with only two pseudolabia (Tkach *et al.*, 2014; Kuzmin, 2013). From the specimens collected, it was possible to identify that the apical morphological characteristics are consistent with the characteristics of the neotropical species, like the presence of six lips uniform in length and shape (Tkach *et al.*, 2014). Morphometric analyzes performed for the *Rhabdias* specimens collected in this study demonstrate morphological characters of the apical region and morphometrics different from the species recorded for Brazil, which led us to the implementation of molecular analyzes for the certification of a possible new species for the genus and a later description (Figs. 7a and 7b).

Species of the genus *Cosmocerca* are widely distributed throughout all continents. The species *Cosmocerca parva* has a large distribution throughout Central and South America, thus having morphological and morphometric variations regarding body length, width and number of plectanas (5 – 7) (Rizvi *et al.*, 2011). According to González and Hamann 2011 these variations occur according to the host and sometimes within the same individual. Even though the specimens found in this study present the same morphological characteristics as the ones described by Travassos 1925, the phenotypic plasticity observed is wide and the morphometric characters observed are smaller than those reported in the literature (Bursey *et al.*, 2015) (Table 3), which may be related to the size of the host (Fig. 7c).

There are currently 30 species described for *Cosmocerca*, of which 10 are described for the Neotropical region (Bursey *et al.*, 2015). Peru, Argentina and Brazil are the countries in South America with the highest numbers of infection records of *C. parva* in amphibians, respectively (Santos & Amato, 2013). In Brazil, the records of this helminth are mainly concentrated in the South and Southeast regions, infecting species of Brachycephalidae, Leptodactylidae, Hylodidae, Hylidae, and Bufonidae (Campião *et al.*, 2014).

Oxyascaris oxyascaris was initially described parasitizing the snake *Mastigodryas bifossatus* Raddi, 1820 (= *Drymobius bifossatus*) in Rio de Janeiro (Vicente *et al.*, 1991). Currently, the genus is composed of four other *Oxyascaris* species: *Oxyascaris similis* Travassos, 1920 (= *Pteroxyascaris similis*), *Oxyascaris caudacutus* Freitas, 1958, *Oxyascaris mcdiarmidi* Bursey and Goldberg, 2007 (Bursey & Goldberg, 2007). In Brazil, there are records of *O. oxyascaris* infecting amphibians of the Leptodactylidae family in the South and Southeast regions (Vicente *et al.*, 1991). In the Northeast of Brazil, the records are restricted to the states of Bahia and Pernambuco (Teles *et al.*, 2015). This species is identified by having a mouth with three lips, muscular esophagus followed by a glandular ventricle, equal spines, and gull-wing and caudal wings absent (Vicente *et al.*, 1991) (Figs. 7d and 7e).

Physaloptera are parasites of all classes of terrestrial vertebrates (Anderson, 2000; Gorgani *et al.*, 2013). Currently, the following species have been registered for South America and Brazil, infecting reptiles and mammals: *Physaloptera liophis* Vicente and Santos, 1974, *Physaloptera obtusissima* (= *P. monodens*) Molin, 1860, *Physaloptera tubinambae* Pereira, Alves, Rocha, Lima and Luque,

Table 4. List of helminths related to the species of the genus *Pseudopaludicola*.

Host	Helminths	Locality	References
Pseudopaludicola boliviana	**Trematoda**		
	Catadiscus sp.		
	Haematoloechus sp.		
	Gorgoderina sp.		
	Bursotrema sp.		
	Plagiorchiata sp.		
	Travtrema sp.		
	Echinostomatidae sp.	Corrientes, Argentina	Duré, 2004; González and Hamann, 2012
	Cestoda		
	Eucestoda sp.		
	Nematoda		
	Cosmocerca sp.		
	Cosmocerca podicipinus		
	Acanthocephala		
	centrorhynchus sp.		
Pseudopaludicola falcipes	**Nematoda**	Corrientes, Argentina	González and Hamann, 2004; 2009
	Cosmocerca podicipinus		
Pseudopaludicola pocoto	**Nematoda**		
	Rhabdias sp.		
	Cosmocerca parva		
	Oxyascaris oxyascaris		
	Physaloptera sp.		
	Spiroxys sp.	Ceará, Brazil	Present study
	Brevimulticaecum sp.		
	Unidentified nematode		
	Acanthocephala		
	Cystacanth		

2012, *Physaloptera praeputialis* Linstow, 1889, *Physaloptera lutzi* Cristofaro, Guimarães and Rodrigues, 1976, *Physaloptera retusa* Rudolphi, 1819 and *P. bainae* Pereira, Alves, Rocha, Lima and Luque, 2014 (Ávila & Silva, 2010; Ávila *et al*., 2012; Pereira *et al*., 2014 and Ramos *et al*., 2016). As for amphibians there is a record of infection by Physalopteridae larvae in the municipality of Angicos (RN) in the host *Rhinella granulosa* Spix, 1824 (Madelaire *et al*., in press). The specimens of *Physaloptera* sp. found in this study present a cephalic colarete formed by the cuticle reflected on the lips and having a mouth with two large, lateral, simple, triangular lips, each provided with a variable number of apical teeth and externally with papillae (Vicente *et al*., 1991) (Fig. 7f).

The genus *Spiroxys* is widely distributed throughout the Eurasian Palearctic, North Africa, North America and Neotropical countries (Hasegawa *et al*., 1998; Mascarenhas & Muller, 2015). Two species of the genus are found in Brazil, *Spiroxys contortus* Rudolphi 1819, described for the South and Southeast and

Spiroxys figueiredoi Freitas and Dobbin 1962, with records for the North-Northeast, Southeast and Central-West regions infecting species of chelonians and snakes (Vicente *et al*., 1993; Bernadon *et al*., 2013; Mascarenhas and Muller, 2015 and Viana *et al*., 2016). Species of the genus *Spiroxys* are currently divided into three groups: (a) characterized by the presence of teeth in each lobe of the pseudolabium, (b) with teeth only in the median lobe and finally Roca and García, 2008 proposed a third group (c) that are without teeth, found in the Eastern, Australian and Ethiopian zoogeographic regions (Purwaningsih, 2015). The species that occur in Brazil, *S. contortus* and *S. figuereidoi*, are included in the second group (Mascarenhas & Muller, 2015; Fig. 7g).

Brevimulticaecum species are described occurring in the continents of Africa, America and Oceania (Vieira *et al*., 2010). The genus is characterized by having smooth lips with winged margins and absence of dentigerous furrows, excretory pore located anterior to the nerve ring and ventricle with short appendages (González
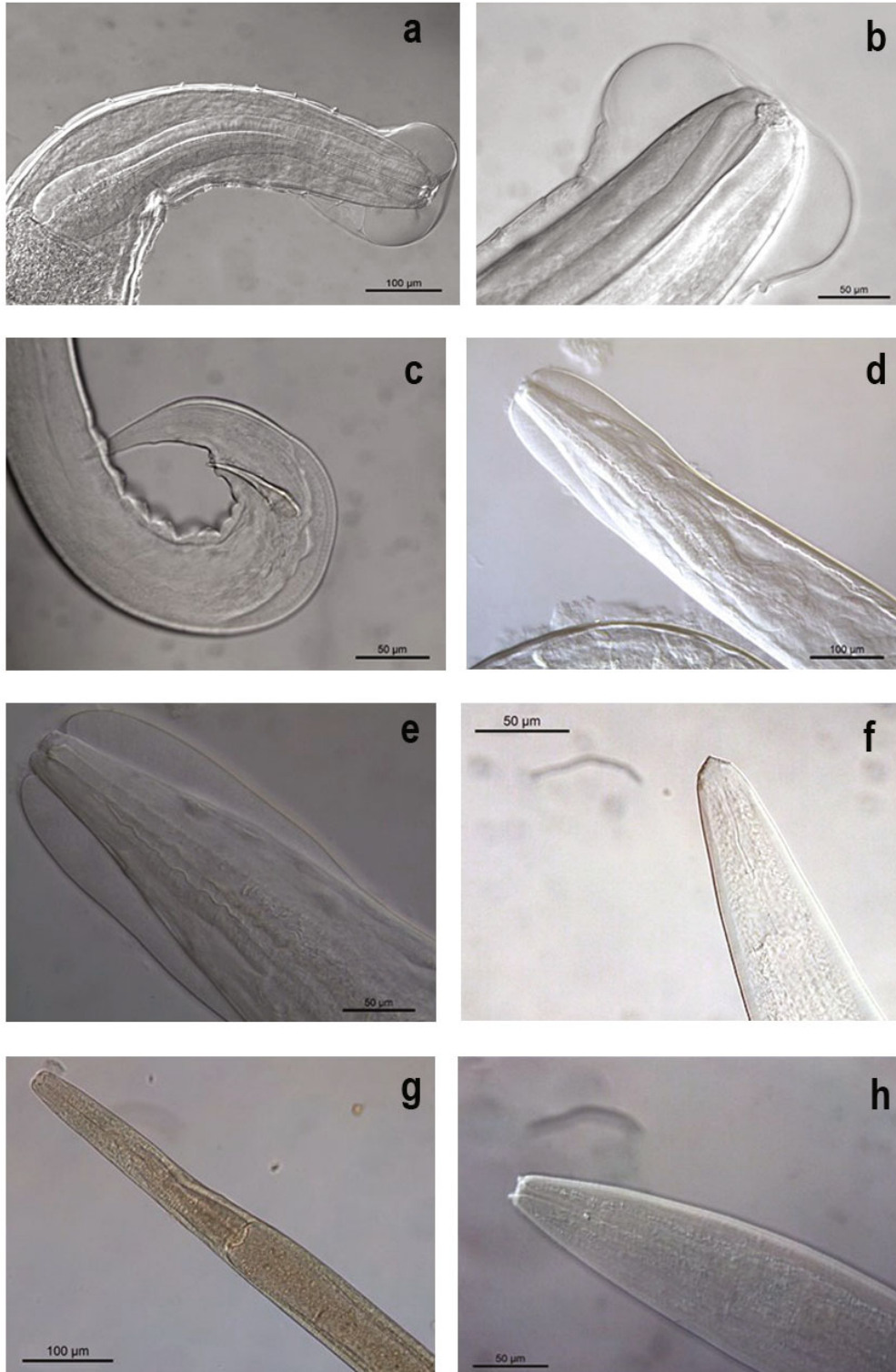
Fig. 7. Photomicrography of the helminth species associated with *Pseudopaludicola pocoto*.
a – anterior region of *Rhabdias* sp. focusing on esophagus and lateral wing; **b** – view of the mouth of *Rhabdias* sp.; **c** – posterior view of the male *Cosmocerca parva*, spicules and plectanas; **d** – anterior view of the male *Oxyascaris oxyascaris*, esophagus and lateral wing; **e** – view of the anterior portion of the male *O. oxyascaris* with emphasis on the mouth and lateral wing; **f** – anterior view of *Physaloptera* sp.; **g** – anterior view of the larva of *Spiroxys* sp.; **h** – anterior view of the larva of *Brevimulcaecum* sp.

& Hamann, 2013). Immature individuals of *Brevimulticaecum* were recorded infecting species such as the Brazilian snake *Bothrops neuwiedi* Wagler in Spix, 1824, the treefrog *Dendropsophus minutus* Peters, 1872 and in the freshwater fishes *Gymnotus carapo* Linnaeus 1758 and *Loricariichthys brunneus* Hancok 1828 (Sprent, 1979; Moravec and Kaiser, 1994; Moravec *et al*., 1997 and Vieira *et al*., 2010) (Fig. 7h).

Accumulation curve based on sampling effort proved to be satisfactory, since the sample reached the asymptote and was representative for sampling the helminth species associated with *P. pocoto*. The helminths richness in *P. pocoto* (S=8) is higher among species of *Pseudopaludicola* (Duré *et al*., 2004; González & Hamann, 2004; 2012). The richness of the helminth infrapopulation of *P. pocoto* varied from one to three species per host, which may be explained by the body size that is a factor influencing the richness and composition of helminth communities (Campião *et al*., 2015). Duré *et al*. 2004 studied the helminth fauna of *P. boliviana* and found a greater species diversity, with representatives of Trematoda (70 %) showing the greatest richness and intensity of infection (Table 4). The cluster analysis indicates that there is more proximity between the helminth communities of *P. boliviana* and *P. falcipes* than between *P. pocoto* and this is due to the fact that *P. boliviana* and *P. falcipes* are sympatric species, and the low similarity between them can be explained by the low sampling of helminths in *P. falcipes* (Fig. 4). In addition, we must also consider that this difference between the community of helminths of *P. pocoto* and the other species of the genus may be due to geographical and environmental differences. The Argentine province of Corrientes, where the species *P. boliviana* was studied, is characterized by wide habitat heterogeneity, many temporary and permanent water bodies, and gleyic arenosols (Duré *et al*., 2004; IUSS, 2015). Thus, aquatic environments can facilitate the occurrence of trematodes, considering that the life cycle of these helminths is heteroxenic, and that in at least one of the phases of parasite transmission is found free in a liquid environment (Travassos, 1950).

Pseudopaludicola pocoto showed a component community mainly composed of nematodes, which may be related to the characteristics of its habitat. The municipality of Aiuaba shows low rainfall levels, annual temperature typical of semiarid climate, and soil composed of arenosols-argillaceous matter (Ipece, 2016). Nematodes are abundant in terrestrial habitats (Ruppert & Barnes, 1996), and the occurrence of these worms in the same habitat as *P. pocoto* enable the encounter of parasites and hosts. The greater abundance of *Rhabdias* sp., *C. parva* and *O. oxyascaris* found in all samplings of this present study suggest that the larvae of these nematode species are present in the habitat throughout the year. This fact can be favored by some habitat characteristics such as high soil humidity, allowing the eggs of these parasites to remain viable in the soil throughout the year giving rise to new larval forms and thus allowing continuity to infection by penetration through the skin in the host (Anderson, 2000; Brito *et al*., 2014).

Although the study period is insufficient to access long-term biocli-

matic predictions on parasite infections of amphibians, the results found herein showed a significant effect of environmental changes on the parasite community, which corroborate other results for the Brazilian semi-arid. Brito *et al.* 2014 found a significant effect of climate variations on the abundance of the helminth community infecting lizards of the family Tropiduridae. In this present study, the environmental changes had an effect on the intensity of infection of *Rhabdias* sp. (Table 2). This lung parasite showed high prevalence in all sampling periods, but was even higher during the rainy season, which may be due to the high humidity in this period. However, climate interaction and rainfall levels did not have effects on the helminth community and species *C. parva* and *O. oxyascaris* in the present study. The investigation of which other variables, like sun radiation, salinity, humidity, and soil pH, are influencing the maintenance of parasite communities is an interesting premise for future studies.

Koprivnikar & Poulin 2009 observed the effects of temperature on cercariae species, in which the temperature was a significant variable increasing the growth rate of these parasites. However, the behavior of these helminths differs in subtropical areas as compared to tropical areas (Choudhury & Dick, 2000; Pizzato *et al*., 2013). For the present study, the temperature did not have a significant influence on the helminth infection rates. Although, a suppressing effect could be observed on the helminth as the temperature increased, even without statistic significance (Table 2).

Environmental changes influence the feeding behavior of hosts (Brito *et al*., 2014), which can affect the infracommunity diversity and infrapopulation abundance of helminths. The migration pattern between the seasons of *C. parva* and *O. oxyascaris* within the infection sites of *P. pocoto* can indicate greater resource availability within the host during the rainy season. This justifies an increase in the occurrence of *C. parva* in the intestinal tract of the host, being found simultaneously with *O. oxyascaris* in the same infection site during that season. However, there might be a greater resource competition in the intestinal tract during the rainy season which may lead *C. parva* to unexplored sites within the host (Esch *et al*., 1990). Still, the occurrence of *C. parva* in the stomach during the dry season can indicate that this species is not a good competitor and by occupying the first organ of the digestive tract this parasite has more chances to obtain food resources (Fig. 5).

The low occurrence of the species *Brevimulticaecum* sp., *Spiroxys* sp. and *Physaloptera* sp. can be explained by the climate conditions of the habitat and also by their life cycle since the three species were recorded during high levels of rainfall. However, the low prevalence and abundance of these nematodes can be related to the fact that species with complex life cycles suffer decrease in population density during trophic transmission (Poulin & Largrue, 2015). Another explanation that justifies the low values of the parasitological descriptors for these genera recorded in this work is the sample effort, which was important to reveal rare species associated with this host. These parasite larvae can be found infecting amphibians that act as second intermediate or paratenic

hosts (Sprent, 1979; Vicente *et al*., 1993; Moravec *et al*., 1997; Goldberg & Bursey, 2007; Goldberg *et al*., 2009 and González & Hamann, 2013). The precise identification of these helminths was not possible because only one specimen of each was found. Investigating and describing the effects of environmental changes on endoparasites associated with members of the family Leptodactylidae can help to elucidate such effects on the dynamics of parasite communities. Besides, inventories of parasite fauna can contribute to studies on host-parasite relation of leptodactylids because they generate new information on the helminths associated with these hosts and provide new guides to identification (Poulin *et al*., 2015). The present study provides new helminth records for *Pseudopaludicola*, and also new records on the range and distribution of some helminth species in the Northeast-Brazilian. The Caatinga biome, with its habitat heterogeneity concentrated in the semiarid, encompasses a diverse fauna of amphibians (Camurugi *et al*., 2010; Andrade *et al*., 2014; Borges-Leite *et al*., 2014 and Cavalcanti *et al*., 2014). Nevertheless, there is still a scarce knowledge of diversity of helminths endoparasites associated with these amphibians.

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