

Rodent helminths in fragmented Atlantic Forest areas in the western region of the state of Paraná

Helminhos de roedores em áreas fragmentadas de Mata Atlântica na região Oeste do Estado do Paraná

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Abstract

Rodents are small mammals that can be parasitized by various helminths. This study aimed to identify and describe the ecological indicators of infection in rodents captured in fragments of the Atlantic Forest in the western region of Paraná State, Brazil. Sixty-eight specimens of five rodent species were collected, necropsied, and inspected in search of helminths. The parasites were stored in 70% ethanol, morphologically identified, and counted for calculation of infection indicators. Fourteen species of helminths and one species of Crustacea were recorded: ten in *Akodon montensis*, four in *Mus musculus*, two in *Thaptomys nigrita*, two in *Oligoryzomys nigripes*, and one in *Euryoryzomys russatus*. The registered species of parasites were: *Rodentolepis akodontis*, *Angiostrongylus* sp., *Protospirura numidica criceticola*, *Trichuris navonae*, *Syphacia alata*, *Syphacia criceti*, *Syphacia evaginata*, *Trichofreitasia lenti*, *Stilestrongylus aculeata*, *Stilestrongylus eta*, *Stilestrongylus graciellae*, *Stilestrongylus franciscanus*, *Stilestrongylus moreli*, *Stilestrongylus* sp., and Pentastomida gen. sp. A positive correlation between the intensity of infection of *T. navonae* and *T. lenti* was observed with the body condition index of the host *A. montensis*. For all species, this study represents a new register of locality, and for eight of them a new host.

Keywords: Rodentia, Nematoda, Cestoda, fragmentation of habitat.

Resumo

Roedores são pequenos mamíferos que podem ser parasitados por uma diversidade de helmintos. Este estudo teve como objetivo identificar e descrever os indicadores ecológicos de infecção por helmintos, em roedores capturados em fragmentos de Mata Atlântica, na região Oeste do Estado do Paraná, Brasil. Sessenta e oito animais foram coletados, necropsiados e inspecionados em busca de helmintos. Os parasitas foram armazenados em etanol 70%, identificados morfologicamente e contados para o cálculo dos indicadores de infecção. Quatorze espécies de helmintos e uma espécie de crustáceo foram registradas: dez em *Akodon montensis*, quatro em *Mus musculus*, duas em *Thaptomys nigrita*, duas em *Oligoryzomys nigripes* e uma em *Euryoryzomys russatus*. As espécies de parasitas registradas foram: *Rodentolepis akodontis*, *Angiostrongylus* sp., *Protospirura numidica criceticola*, *Trichuris navonae*, *Syphacia alata*, *Syphacia criceti*, *Syphacia evaginata*, *Trichofreitasia lenti*, *Stilestrongylus aculeata*, *Stilestrongylus eta*, *Stilestrongylus graciellae*, *Stilestrongylus franciscanus*, *Stilestrongylus moreli*, *Stilestrongylus* sp., e Pentastomida gen. sp. Uma correlação positiva entre a intensidade parasitária de *T. navonae* e *T. lenti* foi observada com o índice de condição corporal do hospedeiro *A. montensis*. Para todas as espécies, este estudo representa um novo registro de localidade, e para oito delas um novo hospedeiro.

Palavras-chave: Rodentia, Nematoda, Cestoda, fragmentação de habitat.

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Introduction

The Atlantic Forest is one of the biomes that suffers most in the world from the processes of habitat loss and fragmentation (Myers et al., 2000; Mittermeier et al., 2005). Most Atlantic Forest remnants are small and surrounded by an anthropic matrix (Ribeiro et al., 2009). This scenario changes the natural structure of populations and metapopulations of small mammals, favoring the emergence of small and isolated populations (Fahrig, 2003), as well as increasing proximity to human and domestic animal populations (Silva et al., 2018).

Rodents are a megadiverse group of mammals, representing 40% to 45% of mammalian species, occupying different biomes (Patton et al., 2015). These animals can act as reservoirs of parasites causing zoonotic diseases (Froeschke & Matthee, 2014), and their eventual proximity to human settlements represent a link between wild and domestic environments, increasing the risk of pathogen transmission to domestic animals and humans (Fundação Nacional de Saúde, 2002; Klimpel et al., 2007; Reperant et al., 2009).

Thus, it is necessary to understand parasitism in rodents living in small fragmented areas of the Atlantic Forest in western Paraná State, Brazil. We investigate the influence of host species, sex, and body condition on helminth parasitic intensity and prevalence, and the influence of the landscape on the structural patterns of the helminth community.

Materials and Methods

Study area

The research was developed in the remaining Atlantic Forest areas in the state of Paraná, Brazil, comprising the cities of Cascavel and Corbélia. These areas are located in the third plateau of the state, in the western region of Paraná, with an altitude varying around 781 m. The climate is humid subtropical, with an average annual temperature of approximately 19 °C. The average maximum temperature in January is 28.6 °C, and in July the average minimum is 11.2 °C, with the occurrence of frosts. The characteristic vegetation is Semideciduous Seasonal Forest (IBGE, 2017).

The remaining Atlantic Forest areas in the region are closely linked to anthropic influences, since the growing demand for food has transformed the surroundings of the small fragments into agricultural landscapes for grain production and dairy farming (Myers et al., 2000). This condition has led to the presence of domestic animals inside and wild animals outside these fragments, since, due to food shortages, rodents have expanded their colonies between and around plantations and peri-domiciliary facilities, and in the home itself (Fundação Nacional de Saúde, 2002; Silva et al., 2018).

Rodent collection

The samples were collected in five small fragments of remaining Atlantic Forest area near the municipalities of Cascavel (24 ° 57 '21 "S, 53 ° 27 '18" O) and Corbélia (24 ° 47 '56 "S, 53 ° 18 '25 "W) in the western region of the state of Paraná. Fragment 1, with an area of 73 ha, Fragment 2, with 96 ha, Fragment 3, covering an area of 28 ha, Fragment 4, with only 11 ha, and Fragment 5, 24 ha of total area (Figure 1).

The blocks are composed of three parallel trapping lines, positioned at the intersection (interface) of the fragment with the adjacent agricultural matrix, 100m away towards the interior of the fragment and the matrix. Each line contained 10 traps set 20m apart from each other, and one pitfall trap, totaling 200m of transept.

Sherman-type (32.4 × 11.7 × 14.2 cm), Tomahawk (29.5 × 11 × 10.5 cm), and pitfall (60 L buckets and canvas barrier 50 cm high by 18 m long) traps were used. The traps were baited with slices of pineapple, a mixture of paçoca (ground peanuts and sugar), oats, grated tuna, and bacon as bait, and were inspected every morning (Mangini & Nicola, 2006).

The sampling occurred simultaneously, over five consecutive nights, in the months of September, November, and December of 2017, and January and February of 2018, corresponding to the initial and late stages of soy cultivation, and April, June, July, and August of 2018, corresponding approximately to the initial and late stages of corn cultivation in the region.

Captured animals were transported to the Laboratory of Metabolism and Human and Animal Physiology of UNIOESTE (Universidade Estadual do Oeste do Paraná - Campus Cascavel), where they were sacrificed by overdose of the volatile anesthetic isoflurane (Brasil, 2012).

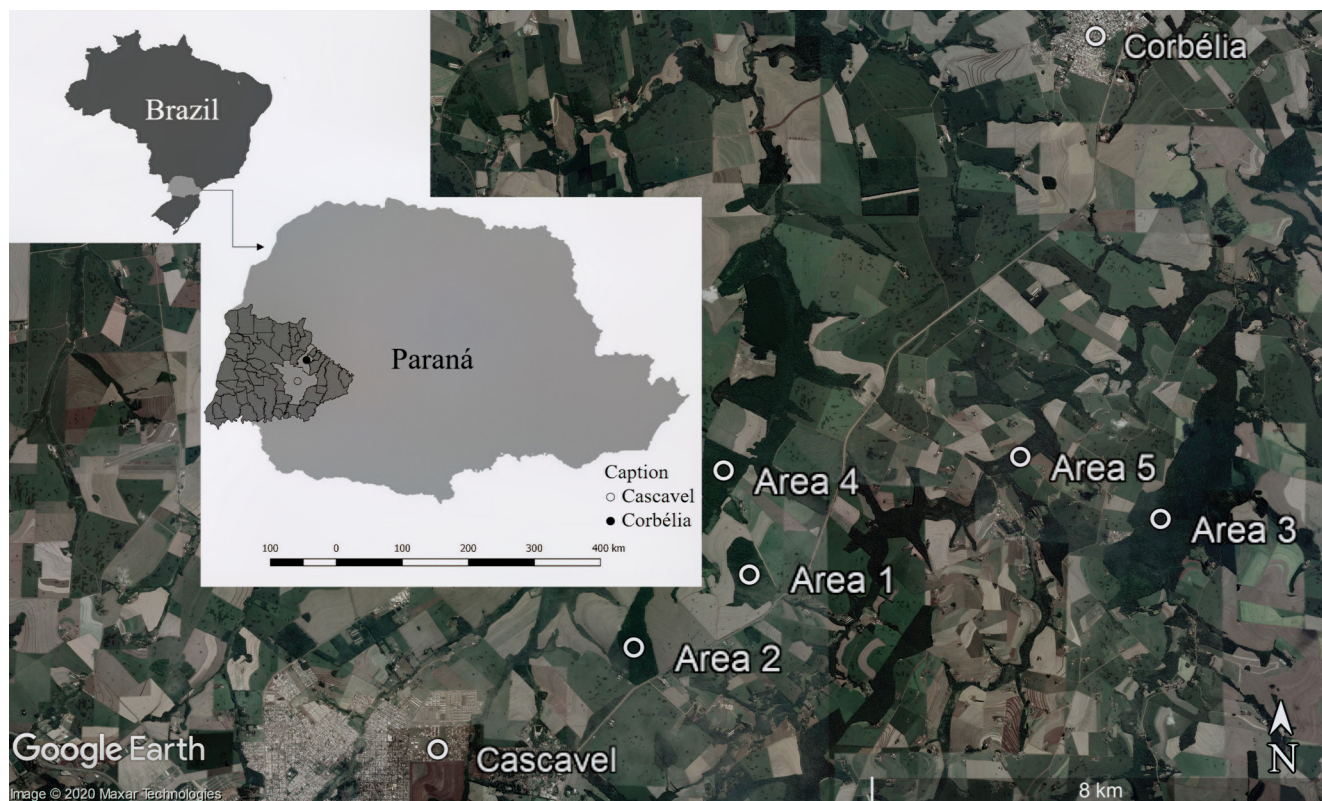


Figure 1. Small rodent sampling locations, from 2017 to 2018, in small Atlantic Forest fragments in western Paraná, Brazil.

Species identification was done through analysis of the guard hairs, by cytogenetics and by analyzing the external and cranial morphology (Quadros & Monteiro Filho 2006; Bonvicino et al., 2008). The skin and skull of the specimens were preserved by taxidermy and skeleton, according to Reis et al. (2010). At necropsy, sex, weight, head-to-tail length, tail length, hind limb length, ear length, and width were recorded for each host, according to Reis et al. (2010).

Each animal was evaluated individually, and all abdominal cavity organs were removed *en bloc*, together with the diaphragm muscle, and stored individually in identified plastic bags at -20°C . The material was then sent to the Laboratory of Parasitic Diseases (LabEPar/UNESP-FCAV), Jaboticabal/SP, for parasitological assessment. The animal captures were authorized by the Instituto Chico Mendes de Conservação da Biodiversidade (SISBIO License 59597-1). All the procedures adopted were approved by the Ethics and Animal Use Committee/UNESP - Jaboticabal/SP (Protocol nº 006060/19) and were performed in accordance with international standards.

Parasitological Necropsy

Helminth collection

Parasitological necropsies were performed in LabEPar, where the anatomical segments of the digestive tract (esophagus, stomach, small intestine, and large intestine), trachea, heart, lung, liver, spleen, and kidneys were sectioned separately in Petri dishes and the contents were analyzed with a stereoscopic microscope for harvesting the helminths, which were later fixed and conserved in 70% ethanol.

Helminth taxonomic identification

The helminths were diaphanized in 80% acetic acid and, if necessary, diaphanized in beech creosote on temporary slides for optical microscopy studies. Taxonomic identification was based on 10 individuals of each sex (dioecious species), 10 adult individuals (monoecious species), or the maximum number of individuals available. To obtain morphometric and morphological characteristics, an Olympus BX-51 microscope with a Q-Color 3 digital camera

was used and the images were processed by Image-Pro Plus 4. Identifications were based on keys such as described by Travassos (1917), Yamaguti (1963), Rêgo (1967), Vicente et al. (1997), Anderson et al. (2009), and additional literature. Vouchers were deposited in the Collection Oswaldo Cruz Institute (CHIOC/Fiocruz) and additional types were kept in the collection of the LabEPar, FCAV/Unesp.

Analysis of the data

After identification and counting, a descriptive analysis of indicators of parasite infection, such as prevalence, mean intensity, and mean abundance, was performed according to Bush et al. (1997). The host body condition index was determined by a comparison of the mass (g) and total length (cm) ($ICC = g/cm$) (Schulte-Hostedde et al., 2005). Only adult specimens were considered for this analysis. Prior to statistical analysis, data distribution was analyzed using the Kolmogorov-Smirnov test. Data with non-normal distributions, were analyzed with non-parametric tests. Fisher's exact test was applied to evaluate the relationship between the sex of the hosts, *A. montensis* and *M. musculus*, and the prevalence of each helminth. The Mann-Whitney test was used to verify the relationship between parasitic intensity and sex in *A. montensis* and *M. musculus* and parasitic intensity and landscape (border and fragment), only in *A. montensis*. Spearman's correlation coefficient was used to verify the relationship between the body condition index and the average parasitic intensity by parasite species. The coefficient of similarity of Jaccard was used to quantify the similarity of helminth communities between the different hosts (*A. montensis*, *O. nigripes*, and *M. musculus*) and the influence of the landscape under the helminth fauna of *A. montensis* (performed only in this host due to the number of samples). All tests were performed using GraphPad Prism 7.04 software with P adjusted to 0.05.

Results

We collected 68 rodents, representing five species of the Rodentia order: 40 *Akodon montensis* (25 Border - 15 Fragment), 20 *Mus musculus* (Plantation), five *Oligoryzomys nigripes* (3 Border - 2 Fragment), two *Thaptomys nigrita* (1 Border - 1 Fragment), and one *Euryoryzomys russatus* (Fragment). Sixty-five rodents were parasitized by at least one helminth species. A total of 4543 helminths were recovered, representing 15 species of three Phyla (Figure 2, Tables 1 and 2). Monoxenous parasites were predominant in all rodents analyzed. Morphometric data on the diagnosed species are listed on Table 3.

All analyzed *A. montensis* were parasitized by helminths. The most frequent species in this host was the trichostrongylid *Stilestrongylus graciellae* (70%, 28/40). The less frequent species were the cestode *Rodentolepis akodontis* (5%) (1/40) parasitizing the small intestine, *Angiostrongylus* sp. (5%) (1/40) parasitizing the lung, and a species of Pentastomida gen. sp. (5%) (1/40), in the form of an encysted nymph in the liver.

Ninety percent of *Mus musculus* (18/20) were parasitized by four species of helminths. The most frequent species, diagnosed in 85% of the rodents, was *Stilestrongylus aculeata* (14/20). *Syphacia evaginata* and *Rodentolepis akodontis* were the least diagnosed species, infecting 30% (6/20) of the analyzed hosts.

Eighty percent of the *Oligoryzomys nigripes* (4/5) were diagnosed with helminths, 40% (2/5) of the rodents were parasitized by *Trichofreitasia lenti* and *Stilestrongylus franciscanus*, parasitizing the bile duct and the small intestine, respectively. Analyzed *Thaptomys nigrita* were parasitized by helminths. The most frequent species, diagnosed in 100% of the animals, was *Stilestrongylus* sp., followed by the oxyurid *Syphacia criceti*, diagnosed in 50% (1/2) of the hosts studied. *Euryoryzomys russatus* was parasitized by the trichostrongylid *Stilestrongylus moreli*.

The highest mean helminth richness was observed in *A. montensis* (total richness 10 and mean richness 2.77). The trichostrongylids presented higher abundance and mean parasitic intensity, followed by the oxyurid species. The less abundant species were Pentastomida gen. sp. (nymph), *Angiostrongylus* sp., *P. numidicola criceticola*, *T. navonae*, and *R. akodontis*.

An intermediate positive correlation was observed between body condition index and mean parasitic intensity involving the nematodes *Trichofreitasia lenti* ($rs^1 = 0.415$; $P = 0.0118$) and *Trichuris navonae* ($rs = 0.477$; $P = 0.0032$) in *A. montensis*. The interactions between: the sex of the hosts and the prevalence of each helminth, was only analyzed in *A. montensis* and *M. musculus*, due to the number of samples; parasitic intensity and landscape (border/fragment) in *A. montensis*; body condition index and total parasitic intensity by species; showed no statistical differences. Jaccard's index showed a low similarity (0.16) between the infra-communities of parasites in the hosts *A. montensis*,

¹ Spearman correlation coefficient

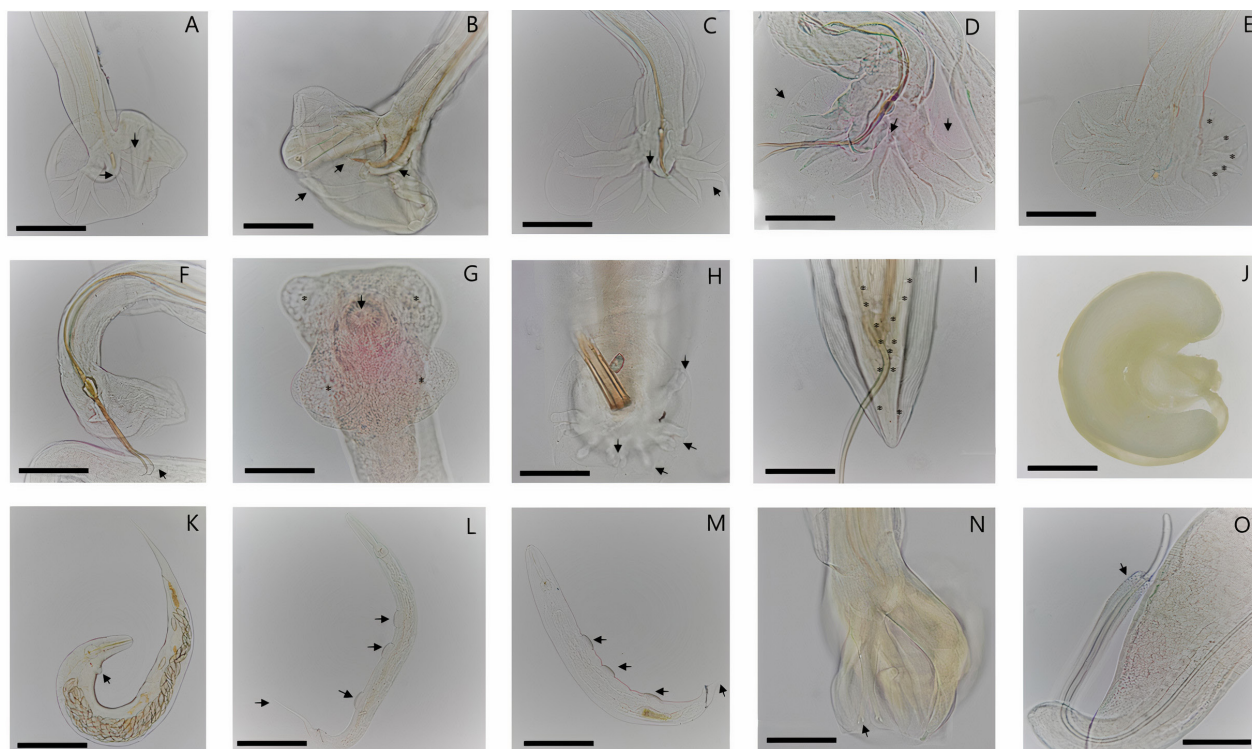


Figure 2. Parasites diagnosed in rodents from small Atlantic Forest fragments in Paraná State, Brazil. **(A)** *Stilestrongylus franciscanus*, externo-dorsal rays are asymmetrical, right external-dorsal ray is thin, arising from proximal third of dorsal ray, the left one is robust, arising at base from dorsal ray, both are longer than the dorsal ray (Scale 50µm); **(B)** *Stilestrongylus* sp. markedly asymmetrical caudal bursa hypertrophied genital cone (Scale 50µm); **(C)** *Stilestrongylus eta*, asymmetric caudal bursa, more developed right lobe, thin ray 8 emerging from the base of the dorsal trunk. (Scale 50µm); **(D)** *Stilestrongylus aculeata*, asymmetric caudal bursa, both externo-dorsal rays originate subsymmetrically at the base of the dorsal part of the trunk, but the origin of the left externo-dorsal ray is more proximal than the right. (Scale 50µm); **(E)** *Stilestrongylus graciellae*, asymmetric caudal bursa with hypertrophied right lobe, type 1-4 pattern, rays 2 appearing first from the trunk than rays 2 to 6 (Scale 50µm); **(F)** *Stilestrongylus moreli*, caudal bursa strongly asymmetric, long and tapered spicules with a curvature in the final portion. (Scale 50µm); **(G)** *Rodentolepis akodontis*, scolex with four suckers without spines, rostellum armed with 20 to 28 hooks (Scale 50µm); **(H)** *Angiostrongylus* sp. fused ventral bursal rays, except at the tips, ventro-lateral rays slightly longer than the ventro-ventral rays, the postero-lateral and medio-lateral rays were fused in the proximal half and separated from the externo-lateral rays after appearance, forming a common trunk, short dorsal bursal ray, ending in tip (Scale 50µm); **(I)** *Protospirura numidica criceticola*, present four pairs of pre-cloacal papillae and one unpaired papilla at the anterior edge of the cloacal opening, two large pairs of sessile post-cloacal papillae and four pairs of small paired papillae located at the tip of the tail (Scale 200µm); **(J)** Pentastomida gen. sp. liver cyst in the nymph stage (Scale 100µm); **(K)** *Syphacia evaginata*, well protruded vulva and the perivulvar region ornamented with a series of cuticular ridges (Scale 200µm); **(L)** *Syphacia alata*, presence of three equidistant ventral mamelons, tail relatively long (Scale 200µm); **(M)** *Syphacia criceti*, presence of mamelons in addition to a spicule, and a gubernaculum with the presence of an accessory hook. (Scale 100µm); **(N)** *Trichofreitasia lenti*, sub-symmetrical caudal bursa, type 2-2-1, dorsal ray divided at mid-length into two branches, each branch divided into two subequal sub-branches, (Scale 50µm); **(O)** *Trichuris navonae*, males without spicular tube, proximal cloacal tube united laterally to distal cloacal tube, cylindrical and spiny spicular sheath (Scale 100µm).

M. musculus, and *O. nigripes*, and a high similarity (0.9) between the landscapes in the rodent *A. montensis*, a test performed only in this host due to the number of samples.

Discussion

The helminthfauna found in this work shows similarities in the composition of the species in relation to those reported in Brazil and Argentina. Nematoda was the most frequent group in this study. Several authors have reported the same findings for other working areas (Cerrado savanna, Pampa grasslands, Caatinga shrubland), and for the same or other small mammal species (Gomes et al., 2003; Navone et al., 2009; Robles, 2011; Simões et al., 2012; Panisse et al., 2017; Hancke & Suárez, 2018; Cardoso et al., 2019).

Table 1. Prevalence (P), mean abundance (A), mean intensity (MI), and range intensity (RI) of rodent parasites in small Atlantic Forest fragments in western Paraná, Brazil.

	Site of infection	<i>Akodon montensis</i>			<i>Oligoryzomys nigripes</i>			<i>Thaptomys nigrita</i>			<i>Euryoryzomys russatus</i>			<i>Mus musculus</i>		
		(n = 40)			(n = 5)			(n = 4)			(n = 1)			(n = 20)		
		P (%)	A	MI (RI)	P (%)	A	MI (RI)	P (%)	A	MI (RI)	P (%)	A	MI (RI)	P (%)	A	MI (RI)
Cestoda																
<i>Rodentolepis akodontis</i> (CHIOC 38790)	SI	5	0.1	2 (2)	-	-	-	-	-	-	-	-	-	30	1.55	5.16 (3-10)
Nematoda																
<i>Angiostrongylus</i> sp. (CHIOC 38791)	L	5	0.1	4 (4)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Protospirura numidica criceticola</i> (CHIOC 38793)	S	10	0.12	1.25 (1-2)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Trichofreitasia lenti</i> (CHIOC 38904)	B	55	5.58	10.1 (1-38)	40	9.2	23 (5-41)	-	-	-	-	-	-	-	-	-
<i>Stilestrongylus aculeata</i> (CHIOC 38795)	SI	30	19.3	64.5 (16-116)	-	-	-	-	-	-	-	-	-	85	23.5	27.7 (1-201)
<i>Stilestrongylus eta</i> (CHIOC 38796)	SI	45	24.1	53.5 (6-204)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Stilestrongylus franciscanus</i> (CHIOC 38797)	SI	-	-	-	40	57	142.5 (120-165)	-	-	-	-	-	-	-	-	-
<i>Stilestrongylus graciellae</i> (CHIOC 38798)	SI	70	29.2	41.7 (2-202)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Stilestrongylus kaaguyporai</i> (CHIOC 38799)	SI	-	-	-	-	-	-	-	-	-	100	13	13 (13)	-	-	-
<i>Stilestrongylus</i> sp. (CHIOC 38794)	SI	-	-	-	-	-	-	100	21.5	21.5 (9-34)	-	-	-	-	-	-
<i>Trichuris navonae</i>	C	37,5	0.82	2.2 (1-6)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Syphacia alata</i> (CHIOC 38800)	C	25	3.1	12.4 (1-27)	-	-	-	-	-	-	-	-	-	-	-	-
<i>Syphacia criceti</i> (CHIOC 38901)	C	-	-	-	-	-	-	50	0.5	1 (1)	-	-	-	50	11.5	23 (6-67)
<i>Syphacia evaginata</i> (CHIOC 38902)	C	-	-	-	-	-	-	-	-	-	-	-	-	30	6.2	20.5 (8-48)
Pentastomida																
Pentastomida gen. sp. (nymph) (CHIOC 38787)	Li	5	0.05	1 (1)	-	-	-	-	-	-	-	-	-	-	-	-

SI, Small intestine; L, Lung; S, Stomach; C, Cecum; Li, Liver; Bile duct, B.

The total helminth richness observed was similar when compared to that reported in other studies conducted in Brazil; Simões et al. (2011) described that *A. montensis* was parasitized by twelve helminth species in the state of Rio de Janeiro. In the state of Santa Catarina; Boullosa et al. (2019) reported six species of helminths in *A. montensis* and two species in *O. nigripes*. Gomes et al. (2003) reported two species of helminths in Rio de Janeiro state, and Cardoso et al. (2018) reported a total richness of six species of parasites in *A. montensis* and two in *O. nigripes* in a preserved area of the Atlantic Forest in the same State. Guimarães et al. (2014) diagnosed three species of helminths in *M. musculus* in the state of Sergipe.

Table 2. Prevalence (P), mean abundance (A) and mean intensity (MI) of each rodent according to intrinsic (sex) and extrinsic (landscape) factors in small fragments of Atlantic Forest in western Paraná, Brazil.

	Parameters											
	Prevalence (%)				Range intensity				Mean abundance			
	Male	Female	Border	Fragment	Male	Female	Border	Fragment	Male	Female	Border	Fragment
<i>Akodon montensis</i>	55	45	62.5	37.5	1	1	1	1	0.55	0.45	0.62	0.37
<i>Rodentolepis akodontis</i>	5	5	7.5	2.5	-	4.5	9	9	0.40	0.5	0.36	0.6
<i>Angiostrongylus</i> sp.	2.5	-	2.5	-	4	-	4	-	0.18	0.22	0.16	0.26
<i>Protospirura numidica criceticola</i>	5	5	7.5	2.5	2.5	2.5	5	5	0.22	0.27	0.2	0.33
<i>Trichofreitasia lenti</i>	37.5	17.5	27.5	27.5	14.13	30.28	19.27	19.27	9.63	11.7	8.48	14.3
<i>Stilestrongylus aculeata</i>	17.5	12.5	17.5	12.5	110.5	154.8	110.5	154.8	35.18	43	30.96	51.6
<i>Stilestrongylus eta</i>	20	25	22.5	22.5	107	107	96.3	120.37	43.77	53.5	38.52	64.2
<i>Trichuris navonae</i>	17.5	20	22.5	15	4.7	4.12	3.6	5.5	1.5	1.83	1.32	2.2
<i>Syphacia alata</i>	12.5	12.5	20	5	26.8	26.8	16.75	67	6.09	7.44	5.36	8.93
Pentastomida gen. sp.	2.5	-	-	2.5	1	-	-	1	0.04	0.05	0.04	0.06
<i>Stilestrongylus gracieae</i>	37.5	30	37.5	30	97.25	97.25	77.8	97.25	53.04	64.83	46.68	77.8
<i>Euryoryzomys russatus</i>	-	100	-	100	-	1	-	1	-	1	-	1
<i>Stilestrongylus kaaguyporai</i>	-	100	-	100	-	13	-	13	-	13	-	13
<i>Thaptomys nigrita</i>	100	0	50	50	1	-	1	1	1	-	0.5	0.5
<i>Stilestrongylus</i> sp.	100	-	50	50	21.5	-	21.5	21.5	21.5	-	43	43
<i>Syphacia criceti</i>	100	-	-	100	-	1	-	1	-	0.5	-	0.5
<i>Oligoryzomys nigripes</i>	100	-	60	40	1	-	1	1	1	-	0.6	
<i>Trichofreitasia lenti</i>	40	-	20	20	23.5	-	47	47	9.4	-	23.5	15.6
<i>Stilestrongylus franciscanus</i>	40	-	20	20	92.5	-	185	185	37	-	92.5	61.6
<i>Mus musculus</i> *	50	35	-	-	1	1	-	-	0.5	0.35	-	-
<i>Syphacia criceti</i>	20	25	-	-	57.5	46	-	-	23	32.8	-	-
<i>Syphacia evaginata</i>	10	5	-	-	61.5	123	-	-	12.3	17.5	-	-
<i>Stilestrongylus aculeata</i>	30	25	-	-	156.7	470	-	-	47	67.1	-	-
<i>Rodentolepis akodontis</i>	10	15	-	-	15.5	10.3	-	-	3.1	4.42	-	-

*Collected only in plantation areas

In Argentina, Navone et al. (2009) reported four species of parasites in *O. nigripes*, Panisse et al. (2017) eight species in *A. montensis*, six species in *O. nigripes*, seven species in *E. russatus* and four species in *T. nigrita*. Panisse et al. (2017) conducted a study in the Misiones region, an area with the same biome and geographically close to the area in

Table 3. Morphometric data of fourteen helminth species identified in small rodents in the Brazilian Atlantic Forest (Measures in millimeters).

	<i>Angiostrongylus</i> sp. ¹		<i>Protoparura numidica criceticola</i> ²		<i>Rodentolepis akodontis</i>		<i>Stilestrongylus aculeata</i>		<i>Stilestrongylus eto</i> ³		<i>Stilestrongylus franciscanus</i>		<i>Stilestrongylus gracielae</i>		<i>Stilestrongylus sp.</i>		<i>Stilestrongylus moreli</i>		<i>Syphacia alata</i> ⁴		<i>Syphacia criceti</i>		<i>Syphacia evaginata</i> ⁵		<i>Trichofreitasia lenti</i>		<i>Trichuris novonae</i> ⁶			
	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀		
Body length	16.7	27.2	24.2	52.3	50	3.17	5.28	2.24	2.24	2.44	2.51	1.76	2.51	2.44	2.76	3.89	5.36	1.29	5.99	1.33	4.27	4.55	6.43	10.82	13.0	24.48				
Body width ¹	0.08	0.10	0.6	0.7	0.81	0.06	0.07	0.05	0.06	0.06	0.05	0.04	0.05	0.06	0.06	0.05	0.06	0.08	0.22	0.11	0.21	0.22	0.10	0.12	0.16	0.21				
Cephalic vesicle	-	-	-	-	-	0.06 x 0.03	0.06 x 0.03	0.04 x 0.02	0.06	0.06	0.04 x 0.03	0.06	0.06	0.06	0.06	0.06	0.06	0.07	-	-	-	-	0.06	0.06	0.06	-	-	-	-	
Nerve ring	-	-	0.3	0.4	-	0.12	0.20	0.08	0.13	0.15	0.10	0.14	0.14	0.12	0.10	0.18	0.21	0.08	0.18	0.10	0.14	0.16	0.21	0.25	0.10	-	-	-	-	
Excretory pore	-	-	0.4	0.5	-	0.26	0.31	0.14	0.25	0.30	0.16	0.20	0.20	0.19	0.18	0.30	0.33	0.32	0.61	0.28	0.41	0.46	0.33	0.36	-	-	-	-	-	
Esophagus ²	0.17	0.19	-	-	-	0.34	0.40	0.30	0.33	0.38	0.26	0.32	0.32	0.32	0.33	0.38	0.44	0.22	0.54	0.21	0.38	0.41	0.52	0.54	-	-	-	-	-	
Portion Muscular Portion	-	-	0.36	0.58	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
Glandular	-	-	3.83	5.63	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
Esophageal bulb	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0.05 x 0.011	0.11	0.06	0.11	0.12	-	-	-	-	-	-	-	
Anterior body length	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	7.71	13.20	-	-	
Posterior body length	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5.40	11.28	-	-	
Spicules	0.25	-	-	-	-	0.37	-	0.32	0.52	-	0.45	-	-	-	-	0.89	-	0.062	-	0.087	-	-	0.23	-	1.77	-	-	-	-	-
Larger spicule	-	-	1.29	-	-	-	-	-	-	-	-	-	-	0.53	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Smaller spicule	-	-	0.46	-	-	-	-	-	-	-	-	-	-	0.51	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Spicular sheath	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1.66	-	-	-	-
Gubernacule	-	-	-	-	-	0.026	-	0.012	-	-	0.012	-	-	0.014	-	0.04	-	-	-	-	-	-	0.04	-	-	-	-	-	-	-
Genital cone	-	-	-	-	-	0.08 x 0.03	-	0.06 x 0.03	0.11	-	0.05 x 0.04	-	-	0.09 x 0.04	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

♂ Males; ♀ Females; ♀ Monoecious; ¹At the esophageal-intestinal junction (mid-part of anterior body in *Trichuris novonae*); ²Esophageal total length; ³Measurements based on mature eggs only; ⁴Based on 3 males and one female; ⁵Based on 4 males and one female; ⁶Based on 9 males; ⁷Based on 3 males and 5 females; ⁸Based on 6 males and 8 females; ⁹Based on 10 females only; ¹⁰Based on 9 males and 6 females.

Table 3. Continued...

	<i>Angiostrongylus</i> sp. ¹		<i>Protospirura numidica critetolae</i> ⁶		<i>Rodentolepis akodontis</i>	<i>Stilestrongylus aculeata</i>	<i>Stilestrongylus eta</i> ⁷	<i>Stilestrongylus franciscanus</i>	<i>Stilestrongylus gracielae</i>	<i>Stilestrongylus sp.</i>	<i>Stilestrongylus moreli</i>	<i>Syphacia alata</i> ⁸	<i>Syphacia criceti</i>	<i>Syphacia evaginata</i> ⁹	<i>Trichofreitasia lenti</i>	<i>Trichuris navonae</i> ¹⁰	
	♂	♀	♂	♀	♀	♂	♂	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀
Anterior mamelon	-	-	-	-	-	-	-	-	-	-	-	0.56	0.07	-	-	-	-
Medial mamelon	-	-	-	-	-	-	-	-	-	-	-	0.52	0.07	-	-	-	-
Posterior mamelon	-	-	-	-	-	-	-	-	-	-	-	0.64	0.08	-	-	-	-
Vulva	-	0.17	-	22	-	0.10	-	0.09	0.07	0.06	0.07	0.85	0.61	0.72	-	0.18	0.34
Ovejector	-	-	-	-	-	0.09	-	0.08	0.07	0.07	-	-	-	-	-	0.09	-
						x		x	x	x						x	
						0.03		0.02	0.02	0.03						0.03	
Anus	-	0.05	-	-	-	0.04	-	0.04	0.03	0.02	-	-	0.9	0.68	0.69	-	0.08
Eggs ³	-	-	-	0.045	0.058	0.07	-	0.06	0.06	0.05	0.07	0.08	0.13	0.07	0.07	-	0.04
				x	x	x		x	x	x	x	x	x	x	x		x
				0.035	0.051	0.04		0.03	0.03	0.03	0.04	0.03	0.04	0.03	0.04		0.02

♂ Males; ♀ Females; ♀ Monoecious; ¹At the esophageal-intestinal junction (mid-part of anterior body in *Trichuris navonae*); ²Esophageal total length; ³Measurements based on mature eggs only; ⁴Based on 3 males and one female; ⁵Based on 4 males and one female; ⁶Based on 9 males; ⁷Based on 3 males and 5 females; ⁸Based on 6 males and 8 females; ⁹Based on 10 females only; ¹⁰Based on 9 males and 6 females.

the present study, showing that ecological similarities can influence parasitism and be associated with the range and composition of the host diet as well as with the environmental characteristics of the analyzed areas, as stated by Poulin (2014).

Helminth richness and composition differed among the five rodent species. The observed and estimated richness indices indicated higher average richness in *A. montensis* (2.77) than in the other host species. Total species richness is dependent on the sample size (Walther et al., 1995) and, in the present study, the sample size was smaller for some rodent species than for *A. montensis*. According to Poulin (2014), host population density is a determinant of the rates of infection of directly transmitted parasites and can be overestimated or underestimated when the sample size is relatively small. Even when a small number of hosts are analyzed, the results help to understand parasite-host dynamics, such as the structure of the parasite community.

Low similarity was observed among hosts in relation to parasite infra-community (Figure 3). The difference in habitat use, and eating habits (Dalmagro & Vieira 2005; Cardoso et al., 2016), may have favored host specificity in some helminth species (Poulin, 2014). Parasite sharing is more likely to occur between biologically, ecologically, and/or phylogenetically related host species (Dallas & Presley 2014; Bellay et al., 2015). In this study, the rodent species are of different genera, this could explain the low similarity of helminth fauna found.

Stilestrongylus aculeata was recovered from rodents of the genus *Akodon* in the state of Rio de Janeiro (Gomes et al., 2003) and in Argentina (Panisse et al., 2017). *Stilestrongylus eta* was recorded in the genus *Akodon* (Durette-Desset & Digiani, 2010), in Rio de Janeiro state. The species *P. numidicola criceticola* has been found in several hosts, including carnivores (Stein et al., 1994) and rodents (Quentin, 1971; Sutton, 1989; Miño, 2008; Simões et al., 2010). *Trichofreitasia lenti* nematodes and *R. akodontis* cestodes were previously found to parasitize *A. montensis* in Argentina (Panisse et al., 2017), and *Oligoryzomys nigripes* in the state of Santa Catarina, Brazil (Boullosa et al., 2019).

Trichuris navonae has been described as parasitizing *A. montensis* in the Misiones Province, Argentina (Robles, 2011), and in Santa Catarina by Boullosa et al. (2019). Digiani & Durette-Desset (2007) reported, for the first time, the species *S. graciellae* parasitizing *Phyllotis* sp. (Sigmodontinae) in the Province of Catamarca, Argentina. *Stilestrongylus franciscanus* is described in the intestine of *Graomys griseoflavus* in the Province of San Luis, Argentina (Digiani & Durette-Desset, 2003). *Stilestrongylus moreli* was reported by Panisse & Digiani, (2018) from the Argentine

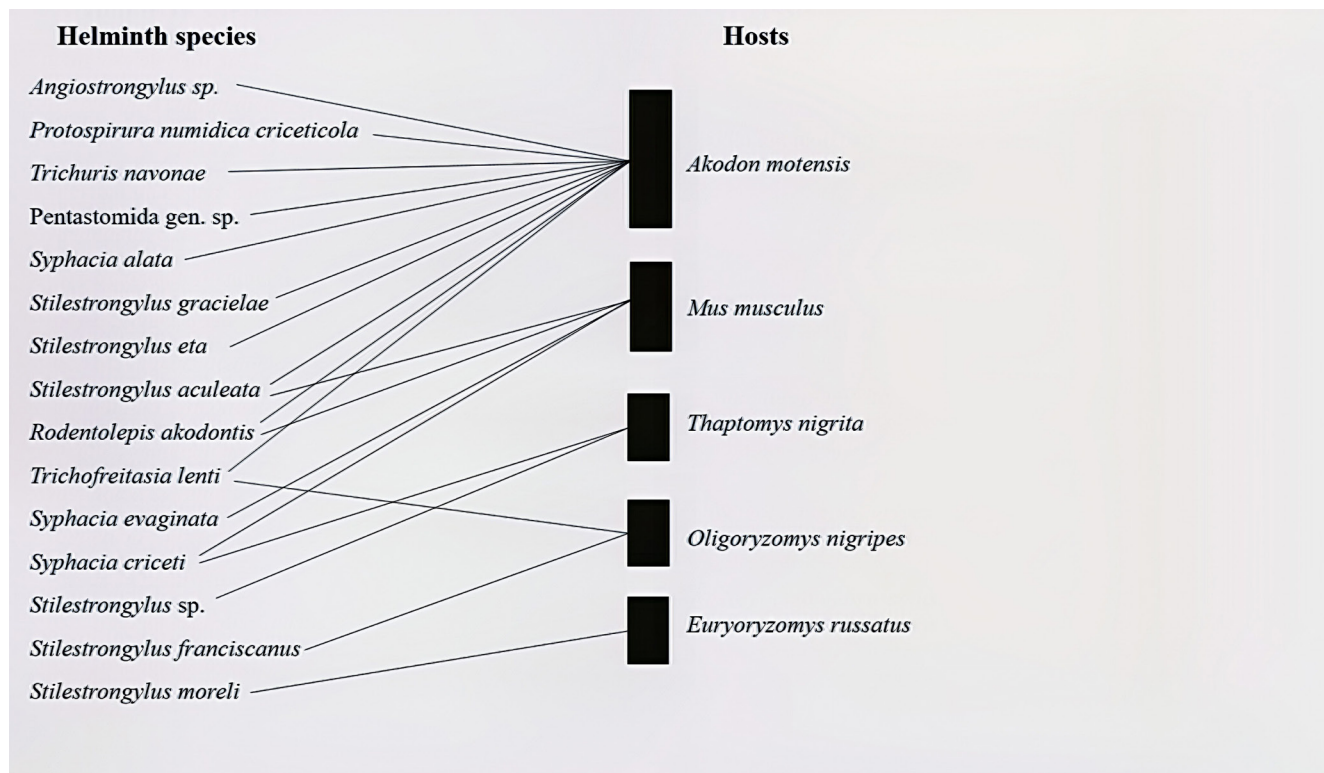


Figure 3. Rodent helminth interaction in five small Atlantic Forest fragments in western Paraná state, Brazil.

Atlantic Forest, in the Misiones province in the rodent *Euryoryzomys russatus*. Species of *Syphacia* are often found parasitizing the cecum of cricetid rodents, mainly sigmodontids (Quentin 1971; Hugot 1988).

Nematodes of the genus *Angiostrongylus* are parasitize rodents and carnivores, residing in the pulmonary or mesenteric arteries of their hosts (Maldonado-Júnior et al., 2012). Two species are pathogenic in humans: *Angiostrongylus cantonensis* causes eosinophilic meningitis or meningoencephalitis, and *Angiostrongylus costaricensis* produces abdominal angiostrongyliasis (Acha & Szyfres, 2003; Maldonado-Júnior et al., 2012; Spratt, 2015). The *Angiostrongylus* sp. specimens that were recovered from the pulmonary arteries of *A. montensis* are morphologically compatible with *A. costaricensis* (Figure 3), but further studies are necessary to clarify this. Unfortunately, we obtained four specimens, and only two of them were not fragmented. As preliminary molecular analyses were not compatible with the generic identification (Benatti et al., unpublished data), additional studies will be performed to confirm the specific identity of the parasite. In an isolate of *A. costaricensis* from Costa Rica, Eamsobhana et al. (2010) showed differences compared to a Brazilian isolate, with an uncorrected p-distance of 11.39%. Jefferies et al. (2009), when comparing isolates of *A. vasorum* from Europe with isolates from South America also showed differences, indicating the possibility of cryptic species.

Pentastomids are parasites of the respiratory system of vertebrates, and most adults are found in the lungs of reptiles, mainly snakes, lizards, and crocodiles (Bush et al., 2001). In rodents, *Porocephalus crotali* is the most reported species, and the nymph stage can be found in the liver, lung, diaphragm, mesentery, and abdominal cavity (Martínez, 1982). The adult form occurs mainly in crotalines (Esslinger, 1962), suggesting that *A. montensis* is involved in the food chain of local snakes.

The helminthfauna of *T. nigrita* has been previously described by Panisse et al. (2017) and Cardoso et al. (2018). These authors reported the occurrence of a new unnamed species of the genus *Stilestrongylus* in Argentina and Brazil. In the present study, the only specimen of *Stilestrongylus* found in *T. nigrita* seems to be the same species found by Panisse et al. (2017), based on the morphological characteristics observed.

Exotic parasite populations in *M. musculus* may originate by transfer from other populations of the same host species or by transfer from other host species in close ecological contact (Tattersall et al., 1994). This could explain the presence of helminths *S. aculeata*, *R. akodontis*, and *S. criceti*, which are most commonly found in wild rodents (Gomes et al., 2003; Panisse et al., 2017). This may be an indication of a spill back event, that is, a parasite, commonly found in native host species, adapting to an exotic host species (Tattersall et al., 1994).

The prevalence and abundance of helminths in wild rodents depend on both extrinsic (temporal, seasonal, and local effects) and intrinsic (age, sex) factors, which interact in various ways to shape the structure of the component community in a given habitat at a specific time (Abu-Madi et al., 2000; Behnke et al., 2005). The age of the host is perhaps the most important of the intrinsic factors, with older or adult animals generally harboring more species of helminths and greater parasitic loading, the so-called cumulative effect (Behnke et al., 1999). In this study, older animals showed greater susceptibility or exposure to *Trichuris navonae* and *Trichofreitasia lenti*, demonstrating that older individuals are more at risk due to the longer exposure time (Behnke et al., 1999).

Sympatric host species generally show differences in feeding strategy and habitat occupation or present distinct activity patterns (Simões et al., 2010). Thus, the establishment of parasitic communities is influenced by the diet of these animals, which, in many species, includes small insects that can act as intermediate hosts of the parasites (Reis et al., 2010). Similarly, the composition and use of the landscape is also a determining factor for host populations (Umetsu & Pardini, 2007; Simões et al., 2010). Some of these factors can be applied to the differences observed in the helminthic community of the analyzed wild rodent population.

This study represents a new location record for all helminth species, extending the geographical distribution of parasites and new host records for *Syphacia alata*, *S. criceti*, *S. evaginata*, *Stilestrongylus graciela*, *S. franciscanus*, *S. moreli*, *S. aculeata*, and *Rodentolepis akodontis*.

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