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ECOSYSTEMS

Diversity and abundance of terrestrial molluscs and their associated nematode fauna in urban kitchen gardens in the city of Rio de Janeiro, Brazil

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Abstract: Terrestrial molluscs can become agricultural pests and transmit parasites, playing an important role in different biological communities. In the present study, we evaluated the diversity and abundance of this group in two horticultural areas in Rio de Janeiro (Manguinhos and Jacarepaguá neighborhoods), as well the presence of parasitic nematodes associated with these molluscs. We collected specimens in the austral spring and summer, with four sites being sampled within each study area, including malabar spinach, sweet potato, chicory greens, and cassava plantations, and one site in an adjacent, non-cultivated area. We collected a total of 522 live mollusc specimens and identified 16 species from 10 different families. The greatest abundance of molluscs was recorded in summer (363) and at Jacarepaguá (309). Overall, 174 (57%) of the 303 specimens analyzed parasitologically were positive for nematodes. Larvae of superfamily Metastrongyloidea, which includes nematodes that are a concern for public health and veterinary medicine, were found parasitizing the slug Sarasinula linguaeformis, in Manguinhos. Our results contribute to understanding of the diversity of terrestrial molluscs occurring in kitchen gardens in the city of Rio de Janeiro, and provide important insights for generating subsidies for health education actions and control of parasitic diseases transmitted by them.

Key words: biodiversity, environmental factors, nematodes, parasitosis, snail, slug.

INTRODUCTION

Most terrestrial gastropods belong to the order Stylommatophora, which includes approximately 15,000 species of snails and slugs, while around 200 species of slugs belong to the family Veronicellidae and less than 100 snails are operculate (Thomé et al. 2006).

Simone (2006) estimated that approximately 2,000 species of terrestrial gastropod occur in Brazil, of which, more than 700 have already been described. These species are highly dependent on their environment, and their activity period is primarily associated with the availability of moisture, which can affect their reproductive functions (Cook 2001, D'Ávila & Bessa 2005). The reduced dispersal capacity of these animals, together with their dependence on the availability of calcium in the soil for shell formation (in snails) and soil humidity, make them valuable models for the understanding of the influence of environmental conditions and impacts on these conditions, and can thus be considered to be important environmental bioindicators (Fernández et al. 2015).

Due to the food preferences and voracity of these animals, which are mostly herbivores,

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they often become agricultural pests, with the potential to generate major impacts on this productive sector (Barker 2002, Thomé et al. 2006, Baronio et al. 2014, Landal et al. 2019, Marchi et al. 2021). Terrestrial molluscs may also present certain risks to the health of humans and domestic animals, given that some species act as intermediate hosts of nematodes that cause parasitosis (Valente et al. 2020). These nematodes include Aelurostrongylus abstrusus (Railliet, 1898) and Angiostrongylus vasorum (Baillet, 1866), which parasitize domestic cats and dogs, respectively (Andrade-Porto et al. 2012). Humans may also be affected through the accidental ingestion of molluscs or food (in particular, leafy vegetables) contaminated with snail mucus containing the larvae of Angiostrongylus cantonensis (Chen, 1935) or Angiostrongylus costaricensis Morera & Céspedes, 1971, which cause eosinophilic meningitis and abdominal angiostrongyliasis, respectively (Rodriguez et al. 2018, Barbosa et al. 2020). The intermediate hosts of these parasites include the Giant African land snail, Achatina *fulica* Bowdich, 1822, which is amply associated with A. cantonensis and A. abstrusus (Da Silva Lima et al. 2020), although other molluscs are also involved (Ramos-de-Souza et al. 2018). In the specific case of A. costaricensis, the principal intermediate hosts in Brazil appear to be slugs of family Veronicellidae. More recently, a slug originating from China, Meghimatium pictum (Stöliczka, 1873), was implicated a case of A. costaricensis infection in humans, in a vineyard infested by the slug (Rodriguez et al. 2018).

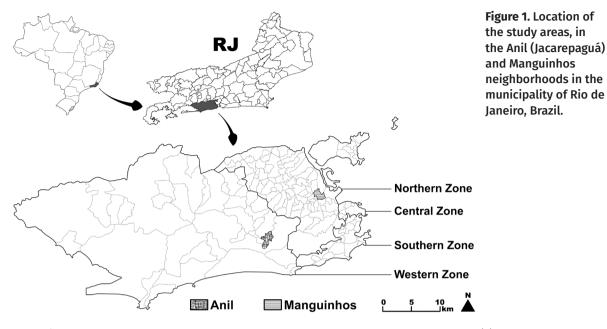
In general, then, terrestrial molluscs, in particular invasive species, have an enormous potential as agricultural pests, including the infestation of commercial and residential gardens, which may also be of epidemiological importance where they provide ideal sites for the development of these animals. In urban areas, these molluscs may also come into contact with definitive hosts (urban rodents) and the etiological agents of the parasitic diseases associated with them (*Angiostrongylus* spp.). The potential occurrence of these parasites in the urban environment, together with the risk of accidental infection of humans through the consumption of inadequately washed vegetables and fruit, make urban kitchen gardens a potential environment for infection by these parasites.

Based on these considerations, the present study investigated the diversity and abundance of terrestrial molluscs in urban kitchen gardens located in the city of Rio de Janeiro, Brazil. We hope that the results of this study will provide valuable guidelines for the control and prevention of the parasitoses transmitted by terrestrial molluscs in this type of environment, providing knowledge that will be useful for local populations, government bodies, and researchers.

MATERIALS AND METHODS

Study area

The present study was conducted in two kitchen gardens in the municipality of Rio de Janeiro, Brazil, one located in neighborhood of Manguinhos (22° 52' 49.3"S, 43° 15' 07.6"W), and other in Anil, in the suburb of Jacarepaguá (22° 57' 52.3"S, 43° 20' 32.6"W), in the northern and western zones of Rio de Janeiro, respectively (see Figure 1). The two vegetable gardens included in this study are part of the "Hortas Cariocas" project, which is run by the Secretary of the Environment of the municipality of Rio de Janeiro. This is a neighborhood-based initiative that supports urban communities.



Sampling

Two field collections were conducted, in November 2019 and February 2020, which coincide with the austral spring and summer, respectively. Four sites were sampled in each study area, represented by three different crops and one non-cultivated site. The size of the sample plots (6 m x 6 m), collection time (10 minutes), and the number of collectors (three) were standardized at each site.

Four different crops were cultivated in the kitchen gardens. Malabar spinach, *Basella alba* Linnaeus (Basellaceae) and sweet potato, *Ipomoea batatas* (Lamarck) (Concolvulaceae), were cultivated in both study areas, while cassava, *Manihot esculenta* Crantz (Euphorbiaceae), was cultivated only at Manguinhos and chicory greens, *Chicorium intybus* Linnaeus (Asteraceae), only at Jacarepaguá. We also evaluated adjacent uncultivated sites within the two study areas, on land used for shaded seedling nurseries and to deposit waste, including wood and tree trunks, dead organic matter, tarpaulins, bricks, and materials used in garden maintenance.

In the vegetable garden located in Manguinhos (area 1), the sampling sites were

numbered from 1 to 4: (1) malabar spinach (22° 52' 51.3" S, 43° 15' 0.79" W); (2) sweet potato (22° 52' 52. 6" S, 43° 15' 0.78" W); (3) uncultivated site (22° 52' 51.4" S, 43° 15' 0.79" W), and (4) cassava (22° 52' 50.7" S, 43° 15' 0.07" W). In Jacarepaguá (area 2) the sites were numbered from 5 to 8: (5) malabar spinach (22° 57' 51.4" S, 43° 20' 32.7" W); (6) sweet potato (22° 57' 53. 2" S, 43° 20' 33.2" W); (7) uncultivated site (22° 57' 52.5" S, 43° 20' 33.4" W), and (8) chicory greens (22° 57' 51.0" S, 43° 20' 30.9" W). The collection of mollusc specimens was authorized by the Brazilian Biodiversity Authorization and Information System, through license number 48371-2, and the specimens were transported to the Laboratory of Malacology at the Institute Oswaldo Cruz/FIOCRUZ in Rio de Janeiro in labeled plastic containers.

Identification of the specimens

The samples were fixed, deposited in the Malacological Collection of the Oswaldo Cruz Institute, CMIOC (catalog numbers CMIOC 12340–12385, CMIOC 12405–12451), and used for taxonomical identification. The taxa were identified based on conchological and anatomical analyses, under a stereomicroscope, supported by the specific taxonomic literature (Simone 2006, Thomé et al. 2006, Rangel et al. 2021) and comparisons with voucher specimens, collected from the city of Rio de Janeiro, deposited in the CMIOC. The specimens were assigned to families based on the classification proposed by Bouchet et al. (2017).

The live specimens of all but two of the species found during the surveys were photographed using a Nikon Canon PowerShot G11 digital camera. In the case of *Allopeas micra* (d'Orbigny, 1835) and *Hutonella bicolor* (Hutton, 1834), it was not possible to obtain a photographic record because the specimens were fixed in the field.

Parasitological analysis

Three hundred and nine alive specimens (303) were used for parasitological analysis. These molluscs were analyzed by artificial digestion, using the Baermann-Moraes technique (Graeff-Teixeira & Morera 1995), to investigate the presence of nematode larvae. Small molluscs (≤ 2mm), such as Beckianum beckianum (Pfeiffer, 1846), Leptinaria unilamellata (d'Orbigny, 1835), Subulina octona (Brüguière, 1792) and Succinea *meridionalis* (d'Orbigny, 1837), were analyzed by pooling specimens (up to 10 specimens). Larger molluscs, including Achatina fulica, Bulimulus tenuissimus (d'Orbigny, 1835), Latipes erinaceus (Colosi, 1921) and Sarasinula linguaeformis (Semper, 1885), were analyzed individually. Other species, including *Bradybaena similaris* (Férussac, 1821), Allopeas gracile (Hutton, 1834), Allopeas micra, Habroconus semenlini (Moricand, 1846), and Ovachlamys fulgens (Gude, 1900), were collected in numbers too small for the parasitological analyses, and the specimens collected were used only for taxonomic identification.

Following artificial digestion, 13 ml of the Baermann-Moraes solution was removed and

examined under an optical microscope for the presence of larval forms with morphological characteristics typical of the superfamily Metastrongyloidea. When encountered, the specimens were stored in saline solution for future molecular identification of the species. The larval forms of other, non-metastrongyloid taxa were also separated and identified to the lowest possible taxonomic level.

Environmental analysis

The ambient temperature (°C) and relative humidity (%) were recorded in the field during the collection the mollusc specimens, using a digital thermo-hygrometer. Soil samples were also collected from each site to determine the physical-chemical parameters of the soil, including total hardness (mg/L CaCO₃), hydrogen potential (pH), conductivity (μ S/cm), and total alkalinity (mg/L CaCO₃). The soil samples were analyzed by Dr. José Augusto Albuquerque dos Santos at the Oswaldo Cruz Laboratory for the Assessment and Promotion of Environmental Health.

Data analysis

The variation in parameters found among the samples was analyzed using the non- parametric Wilcoxon test for pairwise comparisons and the Friedman test for multiple comparisons. Both procedures are used commonly to analyze data in experimental studies. The results are expressed by an index of the difference between means, considering a margin of error and the standard deviation. A $p \le 0.05$ significance level was considered for all analyses. The analyses were run in the GraphPad Prism 5 program (GraphPad Software Inc., San Diego, CA, USA).

The diversity of mollusc species found in the present study was analyzed using the Shannon-Wiener index (H'), Pielou's equitability (J), and Simpson's index of dominance (D) and diversity (1-D), which are the most used in studies of this type (Mendes et al. 2008). These diversity indices are based on the number of species collected (species richness = S) and the abundance of individuals of each species (n). The species composition was compared between study areas using Jaccard's similarity index (Sj), which considers the number of species shared between the areas. These analyses were run in PAST version 2.17c (Hammer et al. 2001).

RESULTS

A total of 760 molluscs were collected in both study areas, including 522 live specimens and 238 shells, from which 16 species were identified, classified into 10 families (Table I, Figures 2 and 3). The 238 shells of the snail species found were not accounted for in the diversity and abundance analyses.

The mean temperatures recorded during the fieldwork were $28.3^{\circ}C(\pm 3.4)$ in the spring and

Table I. Total number of molluscs collected in the urban kitchen gardens in the Manguinhos (MANG) and
Jacarepaguá (JPA) neighborhoods of the city of Rio de Janeiro. X = presence of the species; * = exotic species; ** =
cryptogenic species.

Family	Species	Number of	Collection areas		Number of individuals per collection sites					
		individuals	MANG	JPA	MSpinach	SPotato	OutsideCA	Cas	CGreens	
Achatinidae	Achatina fulica Bowdich, 1822*	84	Х		2	5	50	27	0	
Bradybaenidae	Bradybaena similaris (Férussac, 1821)*	1	Х		0	0	1	0	0	
Bulimulidae	Bulimulus tenuissimus (d'Orbigny, 1835)	38	Х		6	9	13	10	0	
Eucolunidae	Habroconus semenlini (Moricand, 1846)	13		х	4	0	9	0	0	
Heliocarionidae	Ovachlamys fulgens (Gude, 1900)*	2		Х	0	0	2	0	0	
	Allopeas gracile (Hutton, 1834)**	31	Х	х	8	10	4	9	0	
	Allopeas micra (d'Orbigny, 1835)**	9	Х		6	0	0	3	0	
Subulinidae	Beckianum beckianum (Pfeiffer, 1846)**	26	Х	х	9	0	6	0	11	
	Leptinaria unilamellata (d'Orbigny, 1835)*	211		х	53	0	132	0	26	
	Subulina octona (Bruguière, 1792)**	55	Х	х	31	3	10	2	9	
Succineidae	Succinea meridionalis d'Orbigny, 1837	23	Х		6	4	5	8	0	
	Huttonella bicolor (Hutton, 1834)*	1	Х		1	0	0	0	0	
Streptaxidae	Streptartemon cookeanus (Baker, 1914)	2		х	0	0	1	0	1	
Scolodontidae	Tamayoa banghaasi (Thiele, 1927)	3		Х	0	2	0	0	1	
	Latipes erinaceus (Colosi, 1921)	2		Х	1	0	1	0	0	
Veronicellidae	Sarasinula linguaeformis (Semper, 1885)	21	х		0	3	3	15	0	

Abreviations: Manguinhos (MANG), Jacarepaguá (JPA), cultivation of malabar sppinach (MSpinach), cultivation of sweet potato (SPotato), outside cultivation areas (OutsideCA), cultivation of cassava (Cas) and cultivation of chicory greens (CGreens).



Figure 2. Terrestrial molluscs species collected in urban kitchen gardens in the municipality of Rio de Janeiro. a. Achatina fulica, b. Bradybaena similaris, c. Bulimulus tenuissimus, d. Habroconus semenlini, e. Ovachlamys fulgens, f. Succinea meridionalis, g. Latipes erinaceus, h. Sarasinula linguaeformis.

31.1°C (± 0.23) in the summer. Relative humidity was 66% in the spring and 78% in the summer. The mean total hardness of the soil from Manguinhos, that is, the amount of mineral ions (calcium and magnesium), was 45 mg/L, while in Jacarepaguá, it was 55 mg/L. The pH varied little between samples, and the mean was 5.6 for both areas, and alkalinity was 25 mg/L in both areas. Conductivity varied between areas, however, with a mean conductivity of 18.8 μ S/cm in the soil from Manguinhos and 27.8 μ S/cm in that from Jacarepaguá (Table II).

Overall, 159 of the 522 live specimens were collected in the spring and 363 in the summer (Figure 4). At Manguinhos, we collected 46 specimens in the spring and 167 in the summer, for a total of 231 specimens (Figure 4), while at Jacarepaguá, 113 specimens were collected

in the spring and 196 in the summer, with a total of 309 specimens, this being the area with the greatest abundance of molluscs (Figure 4). As the Jaccard index was 18.75%, there was little similarity between areas in the species composition. The number of molluscs collected in the summer (in both areas combined) was significantly higher (Wilcoxon: ** p = 0.0011) than in the spring (Figure 4). A similar seasonal difference was found in both study areas (Figure 4), i.e., Manguinhos (Wilcoxon: ** p = 0.0020) and Jacarepaguá (Wilcoxon: * p = 0.0156), and when the samples are compared between the two study areas (Manguinhos and Jacarepaguá), there was no statistical difference in either the spring (Wilcoxon: p = 0.7349) or the summer (Wilcoxon: p = 0.7720).



Figure 3. Terrestrial molluscs species collected in urban kitchen gardens in the municipality of Rio de Janeiro. a. Allopeas gracile, b. Beckianum beckianum, c. Leptinaria unilamellata, d. Subulina octona, e. Streptartemon cookeanus, f. Tamayoa banghaasi.

The Shannon-Wiener diversity index (H') gives more weight to rare species, while the Pielou (J) evenness indicates the uniformity of the samples. While the Simpson index (1-D) gives greater weight to the most common species, its values were consistent with those of the Shannon index (Table III).

The sample was more diverse in the summer (H' = 2.039) than in the spring (H'= 1.626). Manguinhos (H' = 1.756) had a higher diversity of molluscs overall, in comparison with Jacarepaguá (H' = 1.086). The species were also more evenly distributed in Manguinhos (J = 0.762), and in the summer (J = 0.735), in comparison with the spring (Table III).

The diversity was also calculated for the malabar spinach and sweet potato plantations and the uncultivated sites (Table III), which showed that sweet potato had the greatest diversity of terrestrial molluscs (H' = 1.795) and the best distributed sample (J = 0.922), followed by malabar spinach (H' = 1.754; J = 0.731) and the uncultivated sites (H' = 1.479, J = 0.576). We

compared the diversity of these three types of site, independently of the study area, but found no significant tendency (Friedman: p = 0.0930). We also compared the diversity of the different sites at both Manguinhos (Friedman: p = 0.1585) and Jacarepaguá (Friedman: p = 0.2338), but once again, we found no significant variation among sites.

The molluscs *A. fulica*, *A. gracile*, *B. tenuissimus*, *S. linguaeformis*, *S. octona*, *S. meridionalis*, and *T. banghaasi* (Thiele, 1927) were found in the sweet potato plantations (Table III), and while not especially abundant in this environment, these species richness contributed to the high Shannon index (H'= 1.795). However, the uncultivated sites had the highest species richness, given that 13 of the 16 species identified in the study were found in these sites. These sites also had the greatest abundance of molluscs, with 237 individuals collected, due to presence of large numbers of two species, *A. fulica* (50 specimens) in Manguinhos and *L.*

Parameters	S 1	S2	S 3	S 4	AM	S5	S 6	S 7	S 8	AJ	GA	SD
Soil												
Total hardness (mg/L CaCO³)	50	40	50	40	45	70	60	60	30	55	50	-
Hydrogen potential (pH)	5,8	5,6	5,5	5,4	5,6	5,3	5,4	5,5	5,7	5,5	5,5	-
Total alkalinity (mg/L CaCO³)	25	25	25	25	25	25	25	25	25	25	25	-
Conductivity (µS/cm)	18,1	18,5	18,5	20,1	18,8	18,5	16,7	50,5	25,4	27,8	23,3	-
Climatological												
Temperature (°C) - Spring/2019	33,2	33,2	33,2	26,8	31,6	24	25,1	25,4	25,3	25,0	28,3	±3,4
Temperature (°C) - Summer/2019	30,6	30,3	32,3	31,9	31,3	29	36,9	29,2	28,7	31,0	31,1	±0,2
Relative humidity (%) - Spring/20	59%	59%	59%	81%	65%	86%	87%	87%	77%	84%	66%	-
Ralative humidity (%) - Summer/20	82%	77%	76%	77%	78%	80%	64%	83%	83%	78%	78%	-

Table II. Results of the analysis of the abiotic soil components and climatic data collected in two urban kitchen gardens, in the neighborhoods of Manguinhos and Jacarepaguá, in the city of Rio de Janeiro, Brazil.

Abreviations: sites located in Manguinhos (S1, S2, S3, S4); average of data obtained in Manguinhos (AM); sites localited in Jacarepaguá (S5, S6, S7, S8); average of data obtained in Jacarepaguá (AJ); general average (GA) of the data obtained in Manguinhos and Jacarepaguá; standart deviation (SD)

unilamellata (132 specimens) in Jacarepaguá (Table III).

Overall, 174 molluscs (57%) of the 303 specimens analyzed were positive for nematode larvae (Table IV). Four larval morphotypes were found (Figure 5), which were identified based on morphological characters as: 1. Metastrongyloidea (M1); 2. *Rhabditis* sp. (M2), 3. *Cruzia tentaculata* (Rud, 1817) (M3), and 4. an unidentified sample (M4). Rhabditiform larvae (M2) and M4 were found in *A. fulica*, and M4 in *B. tenuissimus* (Table IV). *Leptinaria unilamellata* was positive for *C. tentaculata* and M4, while *S. linguaerfomis* was parasitized with three larval morphotypes: M1, M2, and M4.

DISCUSSION

Only seven of the 16 mollusc species collected in the present study are considered to be native to Brazil. All the other species are either exotic or cryptogenic, that is, native to other countries and continents or whose origin is uncertain, with

previous records from urban areas in the city of Rio de Janeiro (Alexandre et al. 2017, Rangel et al. 2021). Seven of these species were found only at Manguinhos and six only at Jacarepaguá, while the three other species (A. gracile, B. beckianum, and S. octona) occurred in both areas. These findings are similar to those of Rangel et al. (2021) who recorded 11 species of terrestrial molluscs in anthropized areas around Pedra Branca State Park, in Rio de Janeiro, while Alexandre et al. (2017) found 14 species on the Urca campus of the Federal University of Rio de Janeiro State (UNIRIO), which borders the Carioca Landscape Municipal Natural Park. We recorded a greater abundance of terrestrial molluscs during the summer, where the ambient temperature, relative humidity, and precipitation all tend to increase, in comparison with the spring (Pérez et al. 2008, Almeida 2013).

We considered the malabar spinach plantations to be an "open" cultivation system, in which the sunlight is allowed to reach the soil, which possibly leads to a reduced retention of

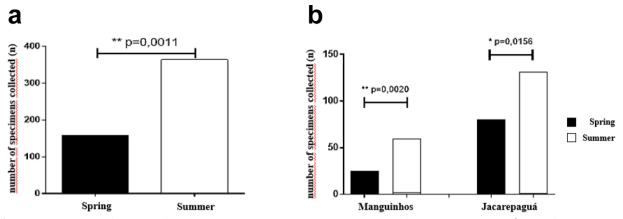


Figure 4. a. Comparative analysis of the total number of molluscs collected in both study areas (Manguinhos and Jacarepaguá) during both field campaigns (Wilcoxon's test). b. Comparative analysis of the total number of molluscs collected in the spring and summer field campaigns, in Manguinhos and Jacarepaguá (Wilcoxon's test).

moisture, in comparison with the sweet potato. This is a climbing plant, with tangled, web-like branches, and while its leaves are well-spaced, their spatial arrangement, together with that of the branches, allows them to partially cover ground, providing shelter for numerous animal species.

We collected a larger number of specimens in the Jacarepaguá vegetable garden, which had a higher index of conductivity than Manguinhos, a parameter that is related to the soil's ability to retain moisture. The relative humidity was also higher in Jacarepaguá, which is also a factor that influences mollusc activity (D'Ávila et al. 2004). D'Ávila & Bessa (2005) and Fischer (2009) demonstrated the influence of different abiotic factors, including moisture, on the life cycle of S. octona and A. fulica, which have a higher growth rate and a larger number of hatched eggs in moister soils. Jacarepaguá also had a greater number of trees and shrubs, as well as a greater availability of calcium in the soil. Calcium carbonate (CaCO3) and pH influence snail development, in particular by contributing to the formation of the shell (Melo et al. 1991, Pacheco et al. 1998). In an experimental study, Hayashi et al. (2005) found that a greater availability of calcium also increases the reproductive

activity of snails, in addition to increasing shell resistance. Other studies, in temperate regions, have also demonstrated the impact of higher calcium and pH levels on the abundance and species richness of terrestrial molluscs (Skeldon et al. 2007, Juřičková et al. 2008).

The area in Jacarepaguá was also more shaded and had a greater amount of organic matter on the ground, in addition to woody debris, tarpaulins, and other materials. Nunes & Santos (2012) observed the influence of luminosity on the distribution of terrestrial molluscs, due to closing of the forest canopy. In the present study, species richness did not vary noticeably between the two study areas, although species composition and abundance were different, which may have been influenced by the local environmental conditions, as also found by Santos & Monteiro (2001), who also used diversity indices to compare study areas.

The Manguinhos kitchen garden had the lowest relative humidity (65–78%) and soil calcium (45 mg/L), in addition to the greatest level of human impact. This garden is less well lit, being located in close proximity to housing and areas with an accumulation of garbage and organic matter, which may influence the presence of invasive species, such as *A. fulica*

Indicators	Spring	Summer	MANG	JPA	MSpinach	SPotato	OutsideCA
Richness (S)	13	16	10	9	11	7	13
Indivíduals (n)	159	363	213	309	127	36	237
Dominance (D)	0,304	0,188	0,226	0,498	0,250	0,188	0,362
Simpson Diversity 1-D	0,695	0,811	0,773	0,501	0,749	0,811	0,637
Shannon Diversity (H)	1,626	2,039	1,756	1,086	1,754	1,795	1,479
Pielou's Equitability (J)	0,634	0,735	0,762	0,494	0,731	0,922	0,576

Table III. Biological diversity of the terrestrial molluscs collected in the urban kitchen gardens in the municipality of Rio de Janeiro as a function of the climatic season (spring or summer) between study areas and among sites.

Abreviations: Manguinhos (MANG), Jacarepaguá (JPA), cultivation of malabar sppinach (MSpinach), cultivation of sweet potato (SPotato) and outside cultivation areas (OutsideCA).

Table IV. Results of the parasitological analysis of the terrestrial molluscs collected from the urban kitchen gardens in Manguinhos and Jacarepaguá, in the city of Rio de Janeiro, Brazil. M1 = Metastrongyloidea; M2 = *Rhabditis* sp.; M3 = *Cruzia tentaculata*; M4 = unidentified specimen; X = presence of the larval morphotype in the mollusc species.

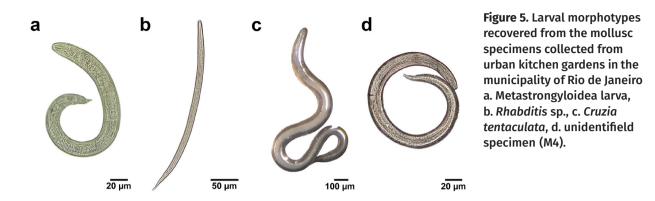
Creation	Nu	mber of individua	Larval morphotypes				
Species	Analyzed (n)	Positive (n)	Positive (%)	M1	M1 M2		M4
Achatina fulica	79	44	56	-	Х	-	Х
Beckianum beckianum	6	0	0	-	-	-	-
Bulimulus tenuissimus	22	13	59	-	-	-	Х
Latipes erinaceus	1	0	0	-	-	-	-
Leptinaria unilamellata	140	110	79	-	-	Х	Х
Sarasinula linguaeformis	19	7	37	Х	Х	-	Х
Subulina octona	22	0	0	-	-	-	-
Succinea meridionalis	14	0	0	-	-	-	-
TOTAL	303	174	57	-	-	-	-

(Colley & Fischer 2009, Silva et al. 2020), which was most abundant species in this study area.

The most abundant species recorded in the present study were the non-native snails *L. unilamellata* (211), *A. fulica* (84), and *S. octona* (55). *Achatina fulica* is considered to be an invasive species that causes a high level of environmental impact (Darrigran et al. 2020). Studies of other non-native species and the damage they can cause are still scarce, although it is known that these species can disrupt the entire ecosystem when introduced into areas where they do not occur naturally (Darrigran et al. 2020). In Jacarepaguá, the number of exotic species (five) was smaller than in Manguinhos (eight), whereas the number of native species was higher in Jacarepaguá (four) than in Manguinhos (three). The environment in the Manguinhos area tends to favor the establishment of molluscs that are associated typically with environmental impacts.

Based on these analyses, and considering the molluscs collected in the two cultivars common to both study areas (malabar spinach and sweet potato), we found a much greater abundance of molluscs in the malabar spinach (127) plantations in comparison with the sweet potato (36). We considered the sweet potato to be a "closed" cultivation, given that the leaves cover a large part of the soil, creating shade, in

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which we expected to find a greater number of both specimens and species. However, we did observe that the irrigation water did not wet the soil completely, with most of it remaining on top of the leaves, without reaching soil.

Despite the diversity and abundance of molluscs found in both study areas, they were not observed feeding on vegetables in either area, which may have occurred because the specimens were collected during the day, when molluscs are at rest (D'Ávilla et al. 2004, Pilate et al. 2017). It is also interesting to note that, prior to the study, municipal employees at both sites reported that they did not usually see molluscs the area, with the exception of some employees at Manguinhos, who commented that they did occasionally encounter *A. fulica*.

The uncultivated sites had the largest numbers of species and also the greatest abundance of molluscs (Table I). At these sites, molluscs were found under debris, wood, stones, fallen tree trunks, and in the layer of organic matter deposited on the ground, which is typical of terrestrial molluscs (Maestrati et al. 2015). Some environments are more suitable for the development of terrestrial molluscs, in particular exotic species, such as areas with piles of garbage, whether organic or formed by inorganic matter, e.g., building materials, which provide shelter for these animals (Raut & Barker 2002, Fischer & Colley 2005, Silva et al. 2020). It is important to note here that anthropogenic changes in the environment can support the emergence of new parasites in known and unknown species, and even in species that had not been previously been recorded as hosts (Webster et al. 2016).

Studies, such as those of Santos & Monteiro (2001) and Klein (1989), have shown how a heterogeneous environment can influence the distribution of species, highlighting the importance of identifying populations. These authors analyzed two areas on Ilha Grande, with different degrees of disturbance. A predominance of herbivorous and generalist species, such as subulinids, was verified in the more anthropogenic area, which led to the conclusion that the composition of the mollusc fauna is related to intensity of anthropogenic impacts and environmental heterogeneity. Klein (1989) mentions that the fragmentation of the vegetation not only alters the habitat, but also the relationships of the species that provide ecological functions in the ecosystem. A heterogeneous environment may thus influence the entire ecosystem, and anthropogenic impacts may accentuate the relationships within the ecological triad - environment, etiological agents, and hosts - creating more favorable conditions for the dissemination of many parasitoses.

One of the nematodes identified in the present study was C. tentaculata, whose definitive host is a marsupial, the opossum (*Didelphis marsupialis*), which has already been recorded infecting A. fulica, Thaumastus taunaisii (Férussac, 1822), and L. erinaceus (Ramos- de-Souza et al. 2021). One of the other nematodes recorded in the present study was a metastrongyloid, a taxon that includes species that cause angiostrongyliasis (Spratt, 2015). This metastrongyloid was found in a specimen of the slug S. linguaeformis in the kitchen garden in Manguinhos. This slug has been found infected naturally with both A. cantonensis and A. costaricensis (Laitano et al. 2001. Caldeira et al. 2007, Carvalho et al. 2012). Angiostrongylus cantonensis has been recorded in the Brazilian state of Rio de Janeiro by Oliveira et al. (2015) and Bechara et al. (2018).

Sarasinula linguaeformis and A. fulica were also been found to be parasitized by nematodes of the genus *Rhabditis*, which is consistent with previous studies (Oliveira et al. 2015, Ramos-de-Souza et al. 2018, Silva et al. 2020). This is the first record of *C. tentaculata* infecting *L. unilamellata*, which was also found parasitized by an unidentified larval morphotype. There are many species of free-living nematodes that sometime appear in the digestive tract of molluscs, including different larval stages, which hampers their taxonomic identification, as observed here and in previous studies (Oliveira et al. 2015).

As parasitic larvae may infect humans accidentally through contact with molluscs, prophylactic measures are necessary to prevent possible parasitosis. Previous research has shown that washing food with sodium hypochlorite kills any viable larvae and guarantees the decontamination of food (Zanini & Graeff-Teixeira 1995). In addition to washing food, it is important to eliminate potential hiding places and avoid the manual gathering of foods. Other potential strategies for the control of molluscs include manual collection, physical traps, and use of chemical and natural baits (Raut & Baker 2002, Vilela & Luengo 2017, De Moura et al. 2018). There is a clear need for a joint initiatives that integrate political, educational, and scientific measures to ensure the effective control of mollusc pests and to prevent diseases that may impact public health and domestic animals.

The scenarios observed in the study areas, highlight the One Health concept, which can be defined as a collaborative effort of multiple disciplines to achieve optimal health for people, animals, and the environment (Webster et al. 2016). The conditions found in the study areas clearly favor the development of the molluscs, demonstrating the multiple connections, so that what affects our environment can also affect both animals and humans, and vice versa. The fact that the molluscs were most abundant and diverse at the uncultivated sites highlights the role of man and society in the formation of the observed scenario.

The diversity of terrestrial mollusc found in the two study areas analyzed here was equivalent to that found in anthropized areas of the city of Rio de Janeiro, including the presence of invasive, exotic and native synanthropic species. Only three species occurred in both study areas, and all three are species of the family Subulinidae, which is common in anthropized areas in the state of Rio de Janeiro. The abundance of the families Achatinidae (Achatina fulica) and Bulimulidae (Bulimulus tenuissimus) at Manguinhos is especially important, given that they were both absent from Jacarepaguá, while the family Subulinidae was represented by four species at this site, including L. unilamellata, which was the most abundant species and was absent from Manguinhos. The families Euconulidae (H. semenlini), Heliocarionidae (O. fulgens), and

Scolodontidae (*Tamayoa banghaasi*) found at Jacarepaguá, were also absent from Manguinhos.

The greater abundance of specimens in the summer, in both study areas, is consistent with previous studies of the influence of humidity on the development of molluscs. The malabar spinach plantation, in which largest numbers of specimens were collected, also appears to form an environment that is favorable to the development of molluscs. The abundance of molluscs found at the uncultivated sites emphasizes the importance of clearing sites around the gardens in order to eliminate breeding sites and thus control infestations. The abundance of non-native species further highlights the level of impact of the study areas, and their susceptibility to invasion by exotic and cryptogenic species, agricultural pests, and potential sources of parasite infection.

The results of the present study demonstrate the importance of studies of this type, as well as the identification and monitoring of areas considered to be of epidemiological interest, such as vegetable gardens, especially those located in urban areas. The urban location of these kitchen gardens may increase the risk of completing the life cycle of metastrongyloid nematodes, in particular species of genus Angiostrongylus, due to the proximity of the definitive hosts in the urban environment. Several of the mollusc species recorded in the present study have been found naturally infected with Angiostrongylus in the literature. In the present study, we highlight the presence of S. linguaeformis infected with metastrongyloid larvae and of A. fulica in the same sitie. This last species is often found infected with nematodes that cause parasitosis (Thiengo et al. 2022). It is recommended the orientation of local population with regard to the potential risks and the prophylactic measures necessary to avoid parasitoses transmitted by molluscs.

In this context, malacological surveys should be implemented in urban horticulture zones, regardless of the size or scale of production. The data collected will be fundamental to the development of effective strategies for the control and prevention of the transmission of molluscan parasites that threaten the health of humans and domestic animals. Any such initiative must integrate a number of different sectors, to ensure the best policies for public health.

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Author contributions

KLO developed the project, selected the kitchen gardens, and collected specimens; conducted the morphological, conchological, and parasitological analyses, the statistical analyses, and wrote the manuscript. PSR participated in the collection of specimens and the parasitological analyses. DD ran the statistical analyses. SRG supervised the development of the project and participated in the writing of the manuscript and the collection of specimens, as well as confirming the taxonomic identification of the mollusc specimens. All the authors discussed the results, reviewed the final manuscript, and are in complete agreement on its content.

