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Spatial distribution and effects of land use and cover on cutaneous leishmaniasis vectors in the municipality of Paracambi, Rio de Janeiro, Brazil

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ABSTRACT

The municipality of Paracambi (Rio de Janeiro, Brazil) reports sporadic cases of American cutaneous leishmaniasis (ACL). Previous studies detected *Nyssomyia intermedia* (Diptera: Psychodidae) as the main vector in the region, but its spatial distribution and the presence of other vector species have not been evaluated. This study aims at filling this knowledge gap, by studying the ecology of sand flies, their spatiotemporal distribution, and correlation with land use/cover. Two campaigns of monthly sand fly collections using light traps and manual captures were conducted in 1992–1994 and 2001–2003. Females were dissected to detect natural *Leishmania* infections. The spatial distribution of sand flies was assessed using kernel density maps. Correlations with land use/cover were evaluated by extracting satellite imagery data around the capture points. A total of 17,232 sand flies from 13 species were captured. Medically important species included *Ny. intermedia*, *Migonemyia migonei*, *Pintomyia fischeri* and *Ny. whitmani*. No *Leishmania*-infected females were detected. Highest densities were detected in the peri-urban areas Cascata and Sabugo, and in rural areas São José and Mutirão. *Ny. intermedia* had statistically significant correlations with pasture and agricultural areas. Present results strengthened that *Ny. intermedia* and *Mg. migonei* are the main local ACL vectors. Correlations with land use evidence the association between ACL and anthropic environmental change.

Introduction

American cutaneous leishmaniasis (ACL) is an anthropozoonosis caused by protozoa of the genus *Leishmania* (Kinetoplastida, Trypanosomatidae), and transmitted from wild, synanthropic, or domestic reservoir hosts to humans through the bite of infected female sand flies (Diptera: Psychodidae: Phlebotominae). In Brazil, the disease is a public health problem that mainly affects vulnerable populations from a socioeconomic point of view, causing cutaneous lesions and/or mucosal lesions in the upper aerodigestive tract. In 2019, 15,484 new cases of ACL were confirmed in the country, with a detection coefficient of 7.37 cases per 100,000 people (Brasil, 2021).

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In Brazil, three species stand out for their major medical and veterinary importance: *Leishmania* (*Leishmania*) *amazonensis*, *L.* (*Viannia*) *guyanensis,* and *L.* (*V.*) *braziliensis* (Brasil, 2017). *Leishmania* (*V.*) *braziliensis* has the widest geographic distribution, multiple ecological interactions, and a large evolutionary history in the Neotropical region (Marzochi et al., 2021). Moreover, *L.* (*V.*) *braziliensis* transmission can establish in peri- and domiciliary environments of peri- and urban areas via adaptation of its associated vectors and host species (Marzochi and Marzochi, 1994).

Approximately 280 species of sand flies (Galati, 2018) occur in Brazil, of which 13 are involved in the transmission of ACL (Brasil, 2017; Rangel et al., 2018b). In the southern and southeastern regions, it has assumed a zooanthroponotic character, with intra- and peridomiciliary transmission associated with secondary or residual forests located in

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the suburbs of urban centers and on occupied slopes in urban regions (Santos et al., 2005; Carvalho et al., 2013; Brasil, 2017; Tanure et al., 2020; Senne et al., 2021). In these circumstances, both humans and dogs, and to a lesser extent, horses, cats and other domestic animals have been infected (Aguillar et al., 1987; Passos et al., 1993; Marzochi and Marzochi, 1994; Schubach et al., 2004; Santos et al., 2005; Figueiredo et al., 2009; Truppel et al., 2014; Lago et al., 2019; Miranda et al., 2019; Salgueiro et al., 2021). In these regions, the species *Nyssomyia intermedia* and *Ny. whitmani* are implicated as main vectors of *L.* (*V.*) *braziliensis* (Costa et al., 2007; Brazil et al., 2015; Rangel et al., 2018b). *Nyssomyia neivai*, *Migonemyia migonei*, *Pintomyia pessoai*, and *Pi. fischeri* are also probably involved in local transmission of *L.* (*V.*) *braziliensis* (Pita-Pereira et al., 2011; Diniz et al., 2014; Brazil et al., 2015)

Rio de Janeiro state is endemic for ACL, despite showing a decreasing trend in prevalence for the last 12 years (SES-RJ, 2021). In 2019, the cases were distributed throughout all the regions of the state, predominantly in the cities of Rio de Janeiro, Paraíba do Sul, Vassouras, São José do Vale do Rio Preto, Teresópolis, Angra dos Reis, and Itaperuna (Miranda et al., 2019; SES-RJ, 2021). *Nyssomyia intermedia* is the main vector, followed by *Mg. migonei* (de Araújo Filho et al., 1981; Rangel et al., 1986; Aguiar et al., 1996; Pita-Pereira et al., 2005; Carvalho et al., 2013, 2014; Senne et al., 2021). Other potential vectors in the state include *Ny. whitmani*, *Bichromomyia flaviscutellata*, *Pi. fischeri,* and *Pi. pessoai* (de Araújo Filho et al., 1981; Rangel et al., 1986, 1984, 1990; Souza et al., 2002; Cardoso et al., 2009).

Paracambi is a municipality of Rio de Janeiro state with sporadic cases of ACL. Approximately 1.3% of cases in the state in the past 10 years were from Paracambi (Santos et al., 2005; Brasil, 2021; SES-RJ, 2021). A study on the prevalence of canine ACL conducted in the 1990s detected between 10% and 27% of infections caused by the *L.* (*V.*) *braziliensis* complex (Santos et al., 2005). In the same study, the presence of *Ny. intermedia* was detected in peri- and domiciliary environments. However, the relative abundance of this vector, *Leishmania* infection rates, and the presence of other potential vector species were not analyzed.

The objective of the present study was to fill the knowledge gap on phlebotomine sand fly fauna in the municipality of Paracambi, focusing on the species of medical importance, in addition to assessing their spatiotemporal distribution and relationships with different ecotopes and types of land use and land cover.

Methods

Study area

The municipality of Paracambi (22º 35’ 22” S, 43º 40’ 43” W) is located on the northwestern limit of the outskirts of the capital of Rio de Janeiro state; it is situated in the Paracambi Mountain Range, with an area of 197 km^2. Its climate is tropical humid, with expressive rainfall, accumulating a mean of 1,400 mm per year. Its annual mean temperature is 24ºC, with a maximum of 27ºC (Natal and Natal, 1987).

Study design

To study the spatiotemporal distribution of sand flies, households were selected based on the following parameters: a) presence of case-indexes of human ACL and domestic animals, mainly dogs; b) organization and occupation of space and land favorable to the occurrence of sand flies; and c) adequate access to the areas.

The first sampling campaign (1992-1994) was aimed at assessing the seasonality of medically relevant sand fly species. Therefore, monthly collections were performed in three selected households at the neighborhoods Cascata and Praça Verde.

In the second sampling campaign, 10 years later (2001-2003), the objective was to evaluate the spatial distribution of sand fly species. In each of the 50 selected households, sand flies were sampled during three nights. The studied localities were: Amapá, BNH, Cabral, Cascata, Centro, Fábrica, Jardim Nova Era, Km 9, Lages, Mutirão, Ponte Coberta, Sabugo, São José, Saudoso and Vila Nova.

All selected households from both study periods were revisited on November 2020 for georeferencing using GPS (Apple® iPhone® SE, My GPS Coordinates software). Coordinates were recorded in the decimal degree format, WGS84 datum, and are available in Table S1.

Collection and processing of sand flies

In each selected household, monthly sand fly captures were conducted in the intra- and peridomiciliary environment with CDC light traps (Sudia and Chamberlain, 1962) and Castro’s manual aspirator (Castro, 1937). The light traps were exposed from 10 p.m. to 6 a.m. (8h of trap/area), installed between 1 and 2 meters from the ground, on the house roof, on tree branches, or in domestic animal shelters (pigsties, chicken coops, and corrals) present in the peridomiciliary environment. Manual captures were conducted from 6 p.m. to 10 p.m. (4h of capture/area) on the houses’ walls (internal and external), animal shelters, and tree trunks, especially at fruit plantations.

Live male specimens were separated in the laboratory and stored in 70% alcohol for usual routine of mounting and species identification, which followed the taxonomic nomenclature of Galati (Galati, 2018), with additional use of the taxonomic keys by Young and Duncan (1994).

The females were dissected over a drop of sterile saline solution on a microscope slide, then mounted between slide and coverslip and observed under light microscopy (400x magnification) to verify possible natural infection by *Leishmania* spp. The head and spermatheca were preserved in 70% alcohol for taxonomic identification of the specimen, following the same method described for the males.

Environmental and cartographic data

Map data from the Mapbiomas project was used to describe the land use and land cover types of the studied region (Souza Junior et al., 2020). These maps include the classification of Landsat satellite images at 30 m spatial resolution for all of Brazil in a time series from 1985 to 2019 (*Mapbiomas Coleção 5*). Images of Paracambi were obtained for the respective years of the sand fly collection campaigns (1992, 1993, 1994, 2001, 2002, and 2003).

The cartographic base of Paracambi municipality was designed in QGIS v. 3.18.3 software (QGIS Development Team, 2021) from data obtained at the Brazilian Institute of Geography and Statistics (IBGE, 2021).

Data analysis

To correct for differences in sampling effort between study periods, all analyses were based on relative abundances, expressed as number of sand flies per trap-night. Total sampling effort was of 35 trap-nights in the first period (1992-1994) and of 81 trap-nights in the second (2001-2003).

Species richness, relative abundances, and total individual counts were assessed for both study periods. Individual frequencies were evaluated per type of capture (manual/light), type of zone of residence (rural/semiurban), and type of ecotope.

For the medically relevant species, differences in abundance between the two study periods were assessed by the student's t test, considering a significance level of 5%. Their seasonality was expressed as the mean monthly relative abundance, using only the results of the first sampling campaign (1992-1994).

Spatial analysis was performed using the data from both study periods. Kernel density maps were generated from the relative abundance values per capture point using a Gaussian function and 200-meter bandwidth, which is in accordance with the average dispersion range of sand flies in forest and peridomestic habitats (Brazil and Brazil, 2018). The data were converted to a logarithmic scale to approximate the normal distribution.

To associate the abundance of medically important sand flies with the different land use and land cover categories, 200-meter buffers were generated at each capture point. In these buffers, the annual percentage cover of the different land use categories was calculated and then associated with the relative abundance of sand flies collected at the same point and year (Figure 1). Pearson's correlation index was used to evaluate this association, considering a significance level of 5%.

Data manipulation and all analyses were performed in R (R Core Team, 2017). For the spatial analyses the packages raster (Hijmans, 2021), rgdal (Bivand et al., 2021), and spatialEco (Evans, 2021) were used. The R code developed for the data analysis is available at GitHub (https://github.com/brunomc-eco/paracambi_sandflies).

Results

A total of 17,232 sand flies were captured, 9,429 between 1992 and 1994, and 7,803 between 2001 and 2003 (Table 1). Thirteen species were

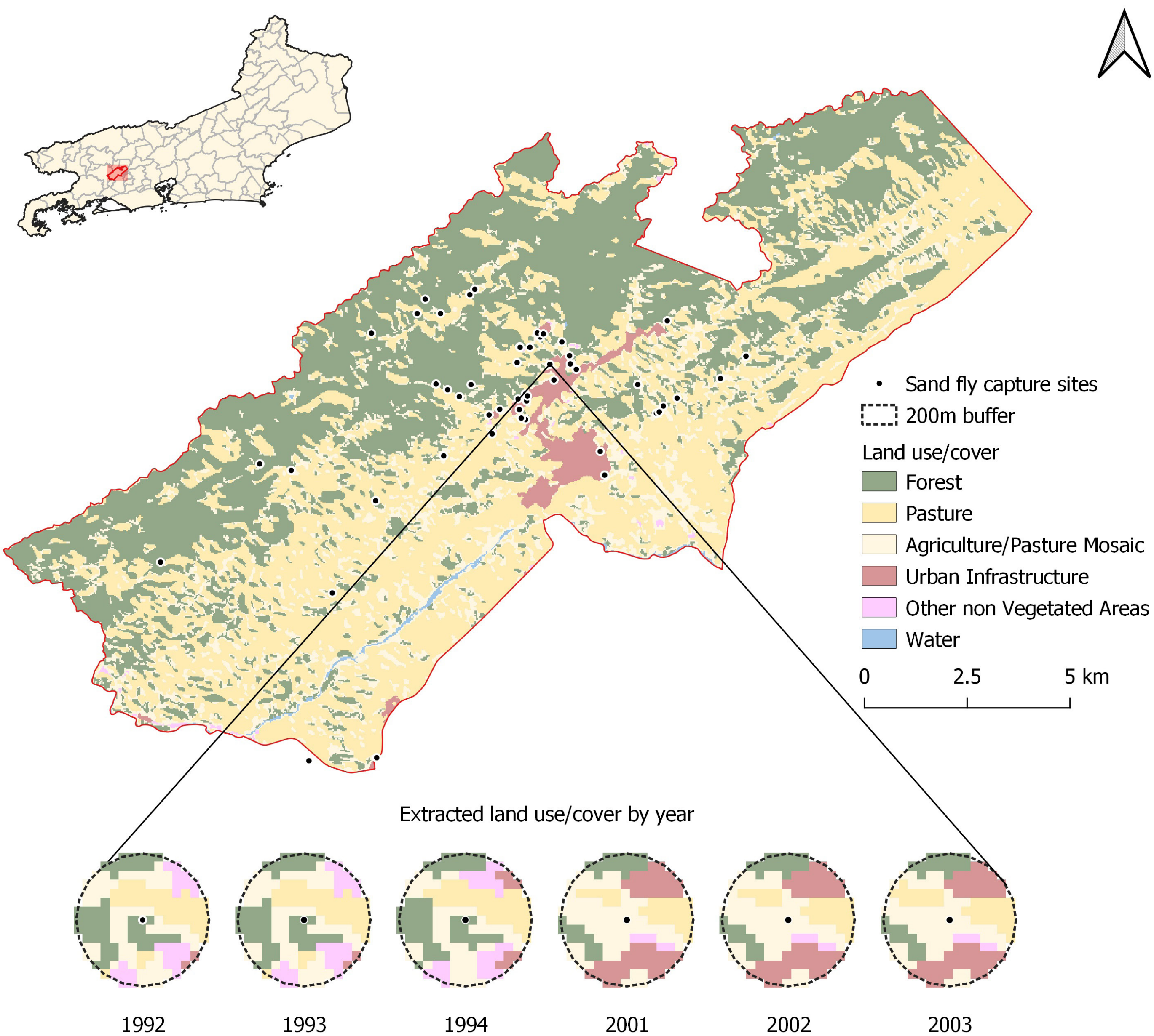

Figure 1 Phlebotomine sand fly capture sites, land use, and land cover in the municipality of Paracambi, Rio de Janeiro State, Brazil. Detail below: example of the extraction of the annual percentages of land use and land cover in a 200-meter buffer in one of the capture sites (1992-1994 and 2001-2003). Spatial resolution of the land cover data is 30 meters (Source: mapbiomas.org).

identified, and the four most abundant with medical relevance were *Ny. intermedia*, *Mg. migonei*, *Pi. fischeri,* and *Ny. whitmani* (Table 1). A total of 879 females of *Ny. intermedia* and 98 of *Mg. migonei* were dissected. However, none of these presented promastigotes of *Leishmania* in their digestive tract contents.

The species *Pi. pessoai*, *Pi. monticola*, and *Psychodopygus hirsutus hirsutus* were captured only in the first period. *Brumptomyia brumpti*, *Br. nitzulescui*, *Evandromyia cortelezzii*, *Micropygomyia schreiberi*, *Psathyromyia pelloni,* and *Sciopemyia sordelli* were detected exclusively in the second period. Despite the differences in species composition and in the absolute number of each species, there was no significant difference between the total numbers captured in the two periods, or in the number of the four most abundant species (Figure S1, Table S2).

Both *Ny. intermedia* and *Mg. migonei* had higher mean relative abundance during summer months, while *Ny. whitmani* peaked during winter (Figure 2). The unusually high values in June for *Ny. intermedia*, *Mg. migonei*, and *Pi. fischeri* were considered outliers, because they were skewed by a single trap-night in a peridomestic banana plantation that yielded approximately 10x more specimens than the average of the remaining trap-nights.

Nyssomyia intermedia, *Mg. migonei,* and *Pi. fischeri* predominated in captures using light traps (56%, 79%, and 67%, respectively), while *Ny. whitmani* had higher frequency in manual captures (52%) (Figure S2). *Nyssomyia intermedia* and *Mg. migonei* were more frequent in the semiurban area compared to the rural area (70% and 57%, Figure S3). *Nyssomyia whitmani* predominated in the rural area (69%, Figure S3). Four species were captured inside the houses: *Ny. intermedia*, *Mg. migonei*, *Ny. whitmani*, *Pi. fischeri,* and also *Pa.* spp. (shannoni complex) (Table 2).

When assessing the spatial distribution of all captured sand flies, higher densities of sand flies were identified in Cascata, São José, Mutirão, and Sabugo neighborhoods (Figure 3). The distribution of sand flies in different ecotopes showed the predominance of species of medical importance in banana plantations, in addition to being found on the outer walls of houses and in domestic animal shelters (Table 2). *Nyssomyia intermedia* was positively correlated with pasture areas and negatively correlated with agriculture/pasture mosaics (Table 3). The remaining tested correlations did not present statistical significance.

Table 1

Species and numbers of sand flies captured by sex and capture period, followed by relative abundances in brackets (specimens per trap-night). Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.

Species	1992-1994			2001-2003			Total
	F	M	F+M	F	M	F+M	
Brumptomyia brumpti	0	0	0	2 (0.02)	2 (0.02)	4 (0.05)	4 (0.03)
Brumptomyia nitzulescui	0	0	0	0	4 (0.05)	4 (0.05)	4 (0.03)
Brumptomyia sp.	1 (0.03)	0	1 (0.03)	0	0	0	1 (0.01)
Evandromyia cortelezzii	0	0	0	0	1 (0.01)	1 (0.01)	1 (0.01)
Micropygomyia schreiberi	0	0	0	0	1 (0.01)	1 (0.01)	1 (0.01)
*Migonemyia migonei**	81 (2.31)	226 (6.46)	307 (8.77)	135 (1.67)	200 (2.47)	335 (4.14)	642 (5.53)
*Nyssomyia intermedia**	4263 (121.80)	4546 (129.89)	8809 (251.69)	2945 (36.36)	4416 (54.52)	7361 (90.88)	16170 (139.40)
*Nyssomyia whitmani**	27 (0.77)	10 (0.29)	37 (1.06)	8 (0.10)	8 (0.10)	16 (0.20)	53 (0.46)
*Pintomyia fischeri**	229 (6.54)	18 (0.51)	247 (7.06)	66 (0.81)	6 (0.07)	72 (0.89)	319 (2.75)
Pintomyia monticola	3 (0.09)	0	3 (0.09)	0	0	0	3 (0.03)
Pintomyia pessoai	4 (0.11)	0	4 (0.11)	0	0	0	4 (0.03)
Psathyromyia pelloni	0	0	0	0	7 (0.09)	7 (0.09)	7 (0.06)
Psathyromyia spp. (shannoni complex)	1 (0.03)	2 (0.06)	3 (0.09)	0	0	0	3 (0.03)
Psychodopygus hirsutus hirsutus	2 (0.06)	0	2 (0.06)	0	0	0	2 (0.02)
Sciopemyia sordellii	0	0	0	2 (0.02)	0	2 (0.02)	2 (0.02)
Unidentified	16 (0.46)	0	16 (0.46)	0	0	0	16 (0.14)
Total	**4627 (132.20)**	**4802 (137.20)**	**9429 (269.40)**	**3159 (39.00)**	**4645 (57.35)**	**7804 (96.35)**	**17233 (148.56)**

**Medically relevant species*
F: females; M: males

Table 2

Species and numbers of sand flies captured by environment (peri- and intradomiciliary) and ecotope in both study periods. Frequencies by ecotope in brackets. Paracambi, RJ, 1992-1994 and 2001-2003.

Species	Intradomestic	Peridomestic					
	Internal walls	External walls	Banana plantations	Other trees	Chicken coops	Pigsties	Corrals
Brumptomyia brumpti	-	-	1 (100%)	-	-	-	-
Brumptomyia sp.	-	-	-	1 (100%)	-	-	-
Evandromyia cortelezzii	-	-	-	-	1 (100%)	-	-
*Migonemyia migonei**	99 (22%)	41 (9%)	140 (31%)	71 (16%)	95 (21%)	3 (1%)	-
*Nyssomyia intermedia**	2503 (21%)	2160 (18%)	5502 (46%)	813 (7%)	762 (6%)	33 (0%)	155 (1%)
*Nyssomyia whitmani**	11 (23%)	3 (6%)	23 (48%)	6 (12%)	3 (6%)	-	2 (4%)
*Pintomyia fischeri**	38 (13%)	37 (12%)	127 (42%)	48 (16%)	49 (16%)	-	2 (1%)
Pintomyia monticola	-	-	3 (100%)	-	-	-	-
Pintomyia pessoai	-	1 (25%)	1 (25%)	2 (50%)	-	-	-
Psathyromyia pelloni	-	-	-	-	6 (100%)	-	-
Psathyromyia spp. (shannoni complex)	2 (67%)	-	1 (33%)	-	-	-	-
Psychodopygus hirsutus hirsutus	-	-	2 (100%)	-	-	-	-
Sciopemyia sordellii	-	-	-	-	1 (100%)	-	-

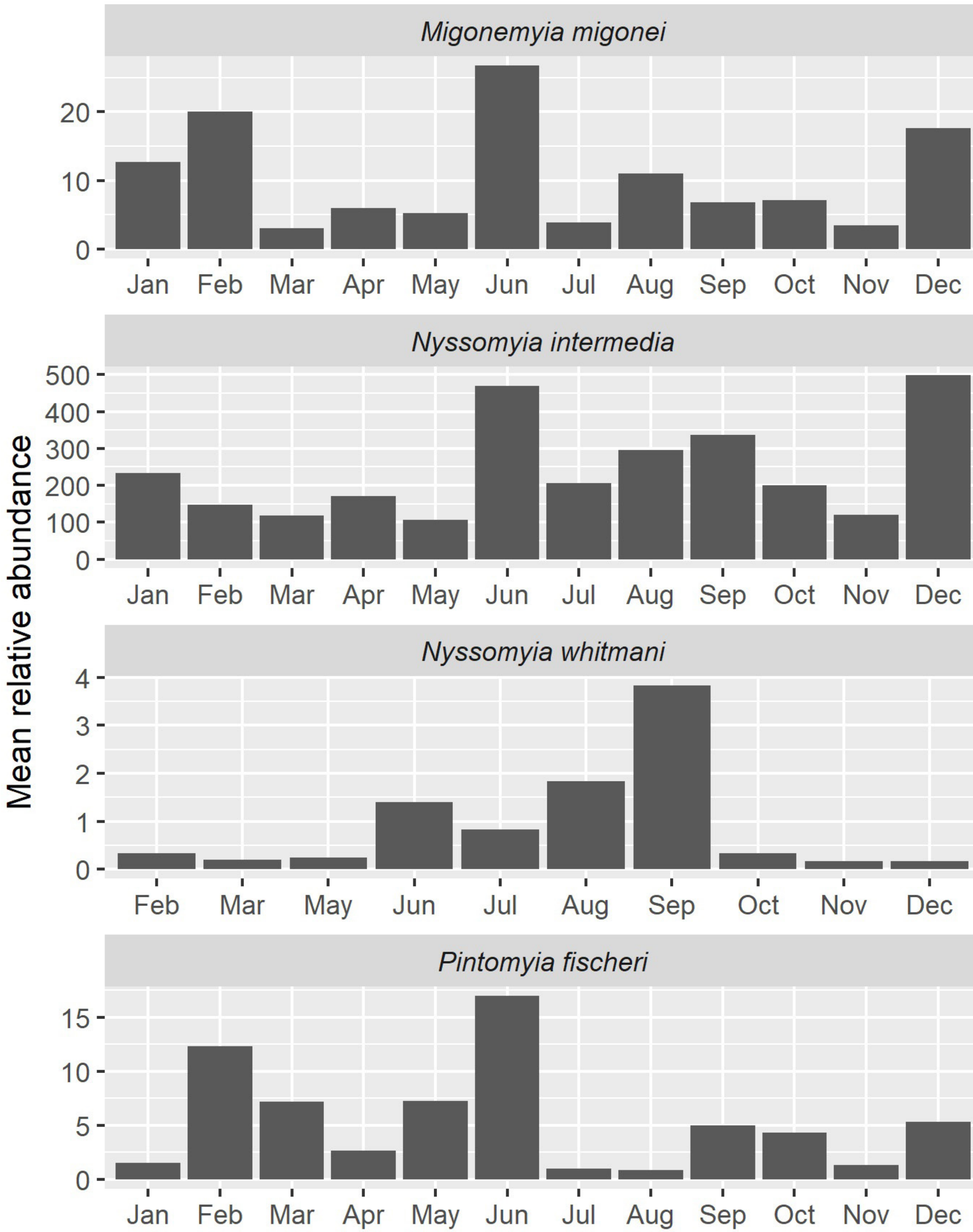

Figure 2 Monthly mean relative abundance of medically relevant sand fly species. Please note the scale difference in the Y axis. Paracambi, RJ, Brazil, 1992-1994.

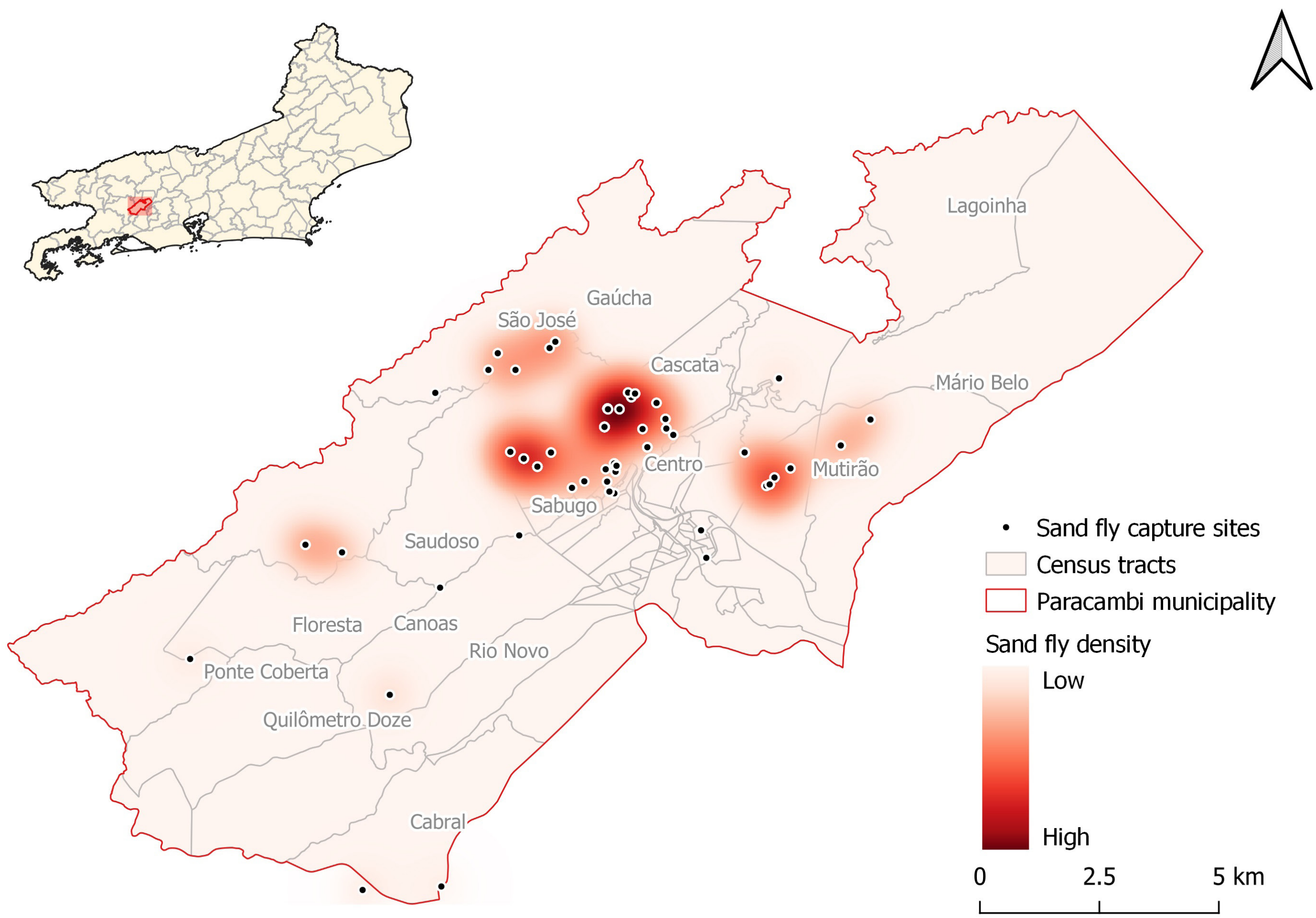

Figure 3 Kernel density map of the total number of sand flies captured in Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.

Table 3
Correlations between the most abundant sand fly species and land use/cover categories, extracted from satellite images of the respective sampling years, in both study periods. Significant correlations are shown in bold. Paracambi, RJ, 1992-1994 and 2001-2003.

Species	Forest	Pasture	Agriculture/Pasture	Non Vegetated	Urban
Nyssomyia intermedia	0.058	**0.325** (.)	**-0.369** (*)	-0.102	-0.192
Nyssomyia whitmani	-0.193	0.097	0.128	0.342	*NA*
Migonemyia migonei	0.413	-0.072	-0.374	-0.198	-0.166
Pintomyia fischeri	0.641	-0.493	-0.158	-0.243	*NA*

Significance codes (***) 0; (**) 0.001; (*) 0.01; (.) 0.05
NA: species was not captured in this land use/cover category

Discussion

The present study detected six new records of sand fly species for the municipality of Paracambi: *Br. brumpti*, *Br. nitzulescui*, *Ev. cortelezzii*, *Mi. schreiberi*, *Pa. pelloni*, and *Sc. sordellii*, in comparison with the pre-existing literature (Santos et al., 2005; Carvalho et al., 2014). All these species have been previously detected in Rio de Janeiro state (Carvalho et al., 2014) and are commonly found in the Atlantic Forest biome, including the neighboring cities of Seropédica (Cardoso et al., 2009; Senne et al., 2021), Itaguaí (Aguiar et al., 1996) and Nova Iguaçu (Oliveira-Neto et al., 1988).

Even with a ten-year interval between the two capture periods, the absence of statistically significant differences in the abundance of species indicates a stability in the sand fly population in the municipality. Species richness was slightly different between the two periods, with a predominance of *Ny. intermedia* and *Ny. migonei* found in all captures. Similar seasonal patterns for *Ny. intermedia* and *Ny. whitmani* were previously detected in Rio de Janeiro state, with the former peaking in the hottest months of the year, and the latter in the coldest months (Souza et al., 2002). In parallel with entomological monitoring in the period 1992–2003, active searches for new human cases of ACL were conducted, when 44 autochthonous cases were reported (Santos GP, Personal communication). For the period 2001–2003, the Ministry of Health Information System on Diseases of Compulsory Declaration (SINAN) reported only seven cases, which suggests underreporting of ACL (Maia-Elkhoury et al., 2007; Brasil, 2021) or a reflection of the movement of the population to reference centers in the city of Rio de Janeiro (Miranda et al., 2019; Salgueiro et al., 2021).

The highest densities of sand flies, as indicated by the kernel map, were detected in Cascata, São José, Mutirão, and Sabugo neighborhoods. One limitation of the kernel density estimator in this study is that it assumes a constant dispersal throughout space. Little is known about the actual dispersal ranges of sand flies in nature, with previous studies having shown that distances vary according to species, sex and environmental features, such as land cover (Brazil and Brazil, 2018). Capture-mark-recapture studies have detected most adult sand flies within an approximately 50-100 m radius, as was seen in Panama forests (57 m; Chaniotis et al., 1974), *Pa. shannoni* in Colombian coffee plantations (100 m; Alexander, 1987), *Ny. intermedia* (109 m) and *Ny. neivai* (100 m) in domestic animal shelters of Vale do Ribeira (São Paulo, Brazil; Galati et al., 2009), and *Lu. longipalpis* in Colombian peridomestic sites (50 m; Morrison et al., 1993). Maximum ranges also vary substantially, with recorded distances up to 500 m (Morrison et al., 1993; Galati et al., 2009) and 1.5 km (Doha et al., 1991). Rather than estimating the actual dispersal ranges of captured sand flies, the kernel approach was applied with the aim of comparing vector densities in the different neighborhoods of Paracambi municipality.

Cascata and Sabugo are located in the peri-urban area of the city, which has mixed characteristics with the rural area, such as small remaining forested areas, banana plantations, and shelter for domestic animals, such as dogs, chickens and, more rarely, pigs. On the other hand, São José and Mutirão are more distant from the city center and have typically rural characteristics, with a large area of forest cover, large areas of agriculture and pasture, and the presence of many animals in the peridomiciliary environment. This pattern of spatial distribution of sand flies in peri-urban areas is common, and has been recently observed, also with the kernel method, in the neighboring city of Seropédica (Senne et al., 2021) and in the suburbs of other Brazilian cities (Machado da Silva et al., 2011; Menezes et al., 2015; Ubirajara Filho et al., 2020).

As in most endemic areas of Rio de Janeiro, this study showed a predominance of *Ny. intermedia* at all collection sites, including inside houses, on internal walls (Rangel et al., 1990; Aguiar et al., 1996; Carvalho et al., 2013; Souza et al., 2015; Vieira et al., 2015). The highest frequencies of its specimens was found in banana plantations (46%), besides being present in domestic animal shelters as previously found by other authors (Rangel et al., 1986, 1990; Aguiar et al., 1996; Souza et al., 2002). The detected correlations of this vector with agricultural and pasture areas further evidences its known process of adaptation to human-modified environments (Meneses et al., 2002; Souza et al., 2002; Rangel et al., 1986, 2018b), being able to colonize these habitats and feed on a variety of domestic animal blood sources (Afonso et al., 2005). This species is considered the main vector of ACL in Rio de Janeiro, with proven vector competence, being highly anthropophilic, and with previous records of natural and experimental infection by *L.* (*V.*) *braziliensis* (Rangel et al., 1984, 1992, 2018b; Pita-Pereira et al., 2005; Rocha et al., 2010).

The species *Mg. migonei* was the second most abundant in this study. It is frequently found cohabiting with *Ny. intermedia* in the southeast region of Brazil, occurring in the houses and in domestic animal shelters, being very anthropophilic, besides biting domestic animals, especially dogs and chickens (Rangel et al., 1986, 1990; de Oliveira-Neto et al., 2000; Souza et al., 2002; Carvalho et al., 2013). This vector was found naturally infected with *L.* (*V.*) *braziliensis* in São Paulo (Pessoa and Coutinho, 1949), Ceará (Azevedo et al., 1990b; Queiroz et al., 1994), and Rio de Janeiro (Pita-Pereira et al., 2005; Carvalho et al., 2013). It is also important to note that *Mg. migonei* was also found naturally infected by *L.* (*L.*) *infantum chagasi*, the etiologic agent of visceral leishmaniasis, in São Vicente Férrer, Pernambuco (de Carvalho et al., 2010). Experimental infection studies have demonstrated the ability of *Mg. migonei* to be infected with both species of *Leishmania*, making it a potential vector of both the cutaneous and visceral forms of the leishmaniases (Guimarães et al., 2016; Alexandre et al., 2020).

The finding of *Pi. fischeri* is relevant because this species has recently gained attention in the literature for being naturally infected with *L.* (*L.*) *infantum chagasi* in the city of Embu das Artes, outskirts of São Paulo (Galvis-Ovallos et al., 2017, 2021). The species has an anthropophilic habit (Aguiar et al., 1996) and despite its potential participation in the transmission of visceral leishmaniasis, the species is more frequently cited in the literature as a potential vector of ACL, with a record of natural infection by *Leishmania* (*Viannia*) sp. in the southern region of Brazil (Pita-Pereira et al., 2011). In the present study, *Pi. fischeri* was found most frequently in banana plantations, but also in chicken coops, corrals, and house walls, both external and internal. These findings corroborate previous studies on its occurrence in the peridomicile in areas of ACL transmission (Teodoro et al., 1993; Aguiar et al., 1996, 2014; Pita-Pereira et al., 2011; Senne et al., 2021).

Even though it was found in low abundance, the presence of *Ny. whitmani* is important because it is the most widely distributed ACL vector in Brazil (da Costa et al., 2018; Rangel et al., 2018b). The present study detected this species in various ecotopes of the peridomiciliary environment, and also in internal walls of houses, despite its low relative abundance. It is a species associated with environmental changes, and it is frequently associated with outbreaks of ACL in areas of recent deforestation (Costa et al., 2007; Rangel et al., 2018b). This vector presents wide ecological valence, and it is present in several environment types, showing varied behaviors and vector competence in each focus of transmission. In the Northeast, Southeast, and South regions, *Ny. whitmani* has been detected naturally infected by *L.* (*V.*) *braziliensis* in different localities (Azevedo et al., 1990a; Queiroz et al., 1994; Luz et al., 2000; Carvalho et al., 2008). In the Northern region, the vector was found to be naturally infected with *L.* (*V.*) *shawi* and *L.* (*V.*) *guyanensis* (Shaw et al., 1991; de Souza et al., 2017). In Rio de Janeiro, *Ny. whitmani* is commonly found at low densities (de Oliveira-Neto et al., 2000; Meneses et al., 2002; Souza et al., 2002; Carvalho et al., 2014), but studies of its seasonality in the region have suggested that this vector alternates peaks of abundance with *Ny. intermedia,* causing transmission of *L.* (*V.*) *braziliensis* to be sustained throughout the year by both species (Souza et al., 2002).

Despite being conducted almost twenty years ago, this study contributes to the knowledge of the local sand fly fauna, which is of high relevance for Public Health authorities and the control of leishmaniasis. Future studies including field surveys in Paracambi are recommended to properly assess the current ecological characteristics of the local sand fly fauna, and its relation with the current distribution of land use classes. In the past recent years (2017-2020), four ACL human cases were notified in the municipality, one of them confirmed as autochthonous (SINAN; Brasil, 2021). American visceral leishmaniasis (AVL) was not officially registered in the municipality to date, but there are notifications of cases in nearby cities, such as Vassouras, Barra Mansa, and Volta Redonda (Brasil, 2021; SES-RJ, 2021). An entomological survey, conducted in Paracambi between September 2001 and February 2002, detected the main vector of AVL in Brazil, *Lu. longipalpis* (Santos GP, Personal communication). On that occasion, seven females and three males were captured on the external wall of houses, using Falcão light traps (Falcão, 1981). The record of *Lu. longipalpis*, associated with its absence in the long-term monitoring performed in this study, suggests that this species is present in low densities in the city. Despite being a vector often associated with urbanized areas (Salomón et al., 2015; Rangel et al., 2018a), in the southeastern region of Brazil, *Lu. longipalpis* was found in areas of forest remnants, suggesting its sylvatic habit as a reflection of the low density in local captures (Cutolo et al., 2009;

de Souza Pinto et al., 2012; Carvalho et al., 2013; Rodrigues et al., 2013; Rangel et al., 2018a). Given the occurrence of AVL in neighbor municipalities and the detection of *Lu. longipalpis*, Paracambi is considered a municipality vulnerable and receptive to AVL, according to the stratification guidelines of the Ministry of Health (Brasil, 2014).

Conclusions

In conclusion, the municipality of Paracambi currently presents low endemicity to ACL. However, the presence of medically relevant sand flies in the peridomiciliary environment shows the potential for transmission of *L.* (*V.*) *braziliensis* in the city. In addition, Paracambi can be considered vulnerable and receptive to AVL. This study concluded that *Ny. intermedia* and *Mg. migonei* are likely the main vectors of ACL in Paracambi, especially in peri-urban areas, given their high abundance, congruent spatiotemporal distribution with the disease, and presence inside houses. Even though this study did not find natural *Leishmania* infection in the dissected females, the presence of this parasite in the city is evidenced by previous studies on the prevalence of canine cutaneous leishmaniasis (Santos et al., 2005) and by the notification of human cases of ACL by the Ministry of Health (Brasil, 2021). The detection of correlation between *Ny. intermedia*, agricultural and pasture areas reinforces the transmission pattern associated with anthropogenic environmental changes, which has been associated with the geographical expansion of ACL in the country (Rangel et al., 2014; Brasil, 2017). The technical-scientific subsidies provided by these results will contribute to the development of control actions by the sanitary authorities of the municipality.

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Conflicts of interest

None.

Author contribution statement

GPLS: Conceptualization, formal analysis, investigation, methodology, project administration, visualization, writing (original draft), writing (review & editing). AS: Conceptualization, funding acquisition, project administration, resources, supervision, validation, writing (review & editing). AOS: Conceptualization, funding acquisition, resources, supervision, validation, writing (original draft), writing (review & editing). MCAM: Supervision, validation, writing (review & editing). AKRG: Investigation, writing (review & editing). BMC: Data curation, formal analysis, methodology, validation, writing (original draft), writing (review & editing). All authors read and approved the final version of the manuscript.

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Supplementary material

The following online material is available for this article:

Table S1 - Geographical coordinates of the sand fly capture points. All named localities are from Paracambi municipality, RJ, Brazil, recorded in WGS84 datum.

Table S2 - Results of the Student's t test comparing the number of captured sand flies belonging to medically important species in the two study periods. Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.

Figure S1 - Comparison of the number of captured sand flies belonging to medically important species captured in the two study periods. Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.

Figure S2 - Frequency of sand flies captured by sampling method – light traps or manual captures. Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.

Figure S3 - Frequency of sand flies captured on each municipality zone – rural or semiurban. Paracambi, RJ, Brazil, 1992-1994 and 2001-2003.