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Original Contribution

Effect of Forest Fragmentation on Tick Infestations of Birds and Tick Infection Rates by *Rickettsia* in the Atlantic Forest of Brazil

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Abstract: Habitat loss and modifications affect biodiversity, potentially contributing to outbreaks of infectious diseases. We evaluated if the patch sizeinfragmented areas of Atlantic Forest in southeastern Brazil influences the diversity of forest birds and consequently the prevalence of ticks on birds and the rickettsial infection of these ticks. During 2 years, we collected ticks from birds in 12 sites: four small forest patches (80–140 ha), four large ones (480–1,850 ha), and four forest control areas within the much larger Morro do Diabo State Park ($\sim$36,000 ha). A total of 1,725 birds were captured (81 species, 24 families), from which 223 birds were infested by 2,339 ticks of the genus *Amblyomma*, mostly by the species *A. nodosum*. Bird diversity and richness were higher in larger than smaller forest fragments. The prevalence of ticks on birds was inversely correlated with bird diversity and richness. Among 174 *A. nodosum* tested for rickettsial infection by polymerase chain reaction, 51 were found to be infected by *Rickettsia bellii* or *Rickettsia parkeri*. However, tick infection rates by *Rickettsia* spp. were not statistically different between forest patch sizes. The higher prevalence of ticks on birds in degraded patches might be caused by a dominance of a few generalist bird species in small patches, allowing an easier transmission of parasites among individuals. It could also be related to more favorable microclimatic conditions for the free-living stages of *A. nodosum* in smaller forest fragments.The higher burden of ticks on birds in smaller forest fragments is an important secondary effect of habitat fragmentation, possibly increasing the likelihood of Rickettsia contagion.

Keywords: ticks, birds, rickettsia, Atlantic Forest, fragmentation, biodiversity

INTRODUCTION

Habitat loss and fragmentation are primary causes of biodiversity loss world wide, increasing the risk of species extinction (Fahrig 2003; Fischer and Lindenmayer 2007). Besides this direct consequence, these processes may also trigger other outcomes, such as the emergence of infection diseases (Daszak et al. 2000; Dobson and Foufopoulus 2001; Daszak and Cunningham 2003; Sutherst 2004; Jones et al. 2008). This potentially disrupts an important ecosystem service, the maintenance of a low and balanced level

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of parasites and pathogens in undisturbed ecosystems (Ostfeld and LoGiudice 2003; Keesing et al. 2010).

We attempted to verify the effect of forest fragmentation on bird diversity, the consequential effect of changes in the prevalence of ticks infesting wild birds, and the rickettsial infection in these ticks. Our hypotheses were that: (i) forest fragment size influences bird diversity and richness; (ii) the richness and the diversity of birds, and the abundance of common bird species, influence the prevalence of ticks on birds; (iii) the richness and diversity of birds influence the prevalence of *Rickettsia* infection rates of ticks.

Our study area was the Atlantic Forest landscape, specifically the Pontal do Paranapanema region in southeastern Brazil. The Atlantic Forest is the fourth most threatened biodiversity hotspot in the world (Mittermeier et al. 1999; Myers et al. 2000), with only 11–16% of the original biome remaining (Ribeiro et al. 2009). Not only is the loss of biodiversity critical in this biome, but so is the (re)emergence of diseases provoked by the intense alteration of this forest and the wide interface of people living around the forest.

A fragmented landscape differs from a continuous one by presenting: (i) a larger amount of edge, which increases interactions among adjacent ecosystems (both natural and anthropogenic) and causes biotic and abiotic changes in both directions across the boundary (Murcia 1995); (ii) a lower connectivity, resulting in higher isolation of species populations among fragments (Tischendorf and Fahrig 2000); and (iii) an increased local probability of species extinction (Ferraz et al. 2007). While the first characteristic is a primary factor in the dissemination of diseases among native and domestic animals, and to humans, the latter two are considered the main factors for biodiversity loss (Fahrig 2003; Uezu et al. 2005; Fischer and Lindenmayer 2007), particularly in a landscape with very low habitat cover (e.g., <30%). More specifically, in a poorly connected landscape, patch size is the primary explanatory variable of species richness, abundance, and composition (Andrén 1994; Pardini et al. 2010; Uezu and Metzger 2011).

Habitat loss and fragmentation might contribute to outbreaks of infectious diseases via both direct and indirect mechanisms. They can do so directly by creating favorable conditions for the proliferation and spread of disease vectors or of species hosts that are competent reservoirs for a pathogen. Examples include an increase of malaria vectors (*Anopheles* sp.) because of optimal microclimate conditions in deforested areas (Yasuoka and Levins 2007), and the increase in songbird exposure to West Nile Virus by increasing levels of urbanization (Bradley et al. 2008).

Habitat loss and fragmentation can also contribute to disease outbreaks indirectly through the loss of biodiversity. It has been suggested that biodiversity plays an important role in decreasing the prevalence of infection disease among hosts due to the dilution effect (Ostfeld and Keesing 2000). In undisturbed ecosystems, biodiversity tends to be greater and more evenly distributed. If a community includes both competent and incompetent reservoir hosts, the potential hosts that are poor reservoirs will dilute the chances of the pathogen dissemination. In contrast, in a disturbed community there may be a bias in the species composition toward the dominance of a few generalist species. If these species are competent hosts, pathogens will spread more easily (Ostfeld and Keesing 2000).

The dilution effect was first proposed for Lyme Disease, also a tick-borne disease (Ostfeld and Keesing 2000). Since then, several studies using simulation models, and experimental and empirical data, have revealed details of this mechanism of disease regulation (Ostfeld and Keesing 2000; Ostfeld and LoGiudice 2003; Keesing et al. 2010). More recently, this effect was also confirmed to explain the prevalence of other diseases, such as the West Nile virus (Ezenwa et al. 2006; Swaddle and Calos 2008; Allan et al. 2009), hantavirus (Clay et al. 2009; Suzán et al. 2009), and schistosomiasis (Johnson et al. 2009).

While the last few decades have seen a substantial improvement in our knowledge about the role of biodiversity in disease prevalence, the majority of studies have been in temperate zones and there is a lack of research in tropical areas. This is critical, as it is the tropics where biodiversity concentrates and at the same time where habitat destruction has been most intense, leading to a high risk of emerging disease hotspots (Sutherst 2004; Jones et al. 2008).

Ticks (Acari: Ixodidae) transmit a huge variety of pathogens and are among the most important vectors of diseases affecting humans and livestock (Jongejan and Uilenberg 2004). Some ticks parasitize a wide variety of hosts, while others are extremely selective and feed solely on one host species (Oliver 1989). For many tick species, the adult stages are more selective for larger sized-mammals, while immature stages (larvae and nymphs) parasitize a wide range of small animals, such as reptiles, rodents, and birds (Hoogstraal 1961; Oliver 1989). Birds also play important roles as carriers of infected ectoparasites and can distribute them within and between continents (Hubalek 2004), which is one mechanism responsible for the wide geographic distributions of certain tick-borne pathogens (Anderson et al. 1986; Elfving et al. 2010; Hildebrandt et al. 2010).

In South America, the most important tick-borne zoonotic disease is Rocky Mountain spotted fever (RMSF), which is caused by the bacterium *Rickettsia rickettsii* (Labruna 2009). Bacteria of the genus *Rickettsia* are obligate intracellular organisms that infect invertebrate hosts worldwide (Raoult and Roux 1997; Perlman et al. 2006). RMSF was first recognized in Brazil in 1929, in the state of São Paulo (Dias and Martins 1939). After a relatively quiet period during the 1960s and 1970s, the disease reemerged in the state and became a serious public health problem of much greater magnitude. During the past two decades, approximately 350 laboratory-confirmed cases (case-fatality rate ≈30%) have been reported in southeastern Brazil (Labruna 2009). The bacterium *R. rickettsii* is transmitted in Brazil by the ticks *Amblyomma cajennense* and *Amblyomma aureolatum*, especially in areas of contact between anthropogenic and natural ecosystems. However, other *Amblyomma* ticks seem to be involved in the transmission of other rickettsiae, with several other *Rickettsia* species with suspected or confirmed pathogenicity to humans being recently reported to infect ticks in South America (Labruna 2009; Spolidorio et al. 2010).

MATERIALS AND METHODS

Study Site

'Pontal do Paranapanema' is in the extreme western part of the Atlantic Forest region, one of the most threatened of the global biodiversity hotspots (Myers et al. 2000). It covers an area of ~245,000 ha in the state of São Paulo (22°53′S, 52°09′W, Fig. 1). The process of deforestation in the region is relatively recent, beginning about 50 years ago, but only 17% of the original biome remains in a matrix composed mainly of pastures and sugar cane plantations (Leite 1998). The mean annual precipitation is 1,500 mm. The original forest type is considered to be seasonal semi-deciduous (Oliveira-Filho and Fontes 2000), being shaped by two extreme climatic seasons, a tropical season with intense rain during spring and summer, and a subtropical dry season with physiologic drought caused by low temperatures during autumn–winter (Veloso et al. 1991). An important element in the landscape is the Morro do Diabo State Park, which covers about 36,000 ha, and constitutes the largest Atlantic Forest remnant in the interior of the state of São Paulo (Leite 1998). In the proximity of this park, there are many forest remnants that vary in size from two to 2,000 ha (Ditt 2002).

Landscape Parameters and Study Design

Previous studies in the region considered the whole community of birds from 28 areas (21 forest patches and seven forested control areas inside the Morro do Diabo State Park) and revealed that bird richness and diversity are particularly influenced by forest patch size (Uezu and Metzger 2011). For the present study, we selected 12 sample sites: four small forest patches (80–140 ha; S1, S2, S3, S4), four large forest patches (480–1,850 ha; L1, L2, L3, L4), and four control areas within the Morro do Diabo State Park (~36,000 ha; C1, C2, C3, C4) (Fig. 1). All sample sites were forest with a similar vegetation structure and were a subset of the sample units previously studied (Uezu and Metzger 2011). All sites were within a similar matrix of pasture and sugar cane plantations except for the control sites, which were within the large forested area of Morro do Diabo State Park. Patch sizes were obtained from maps generated from satellite image classification (A. Uezu, unpublished data).

Capture Events

Forest birds were caught every season from December 2004 to December 2006 in each sampled forest area. In total, there were eight capture events per forest area at 3-month intervals. For each capture event, 15 mist nets (12 m long × 2 m wide, 36 mm mesh) were deployed in the forest during the day, starting between 6:30 and 7:30 am depending on the season, totaling 23,688 net-hours. To avoid potentially strong influences from the edge-effect, nets were deployed a minimum distance of 50 m inside from the forest edge. We identified captured birds to the species, banded, and examined them for the presence of ticks, which were all collected for later identification.

During each capture period, collection of free-living ticks was carried out in the same forest patch of bird captures using the standard method of dragging a piece of white flannel (100 cm × 50 cm) along 1,000 m of forest trails as previously described (Ogrzewalska et al. 2009a). For counting purposes, nymphs and adult ticks were counted individually, whereas larvae were counted as the number of clusters (i.e., each larval cluster collected from the environment was considered a single unit).

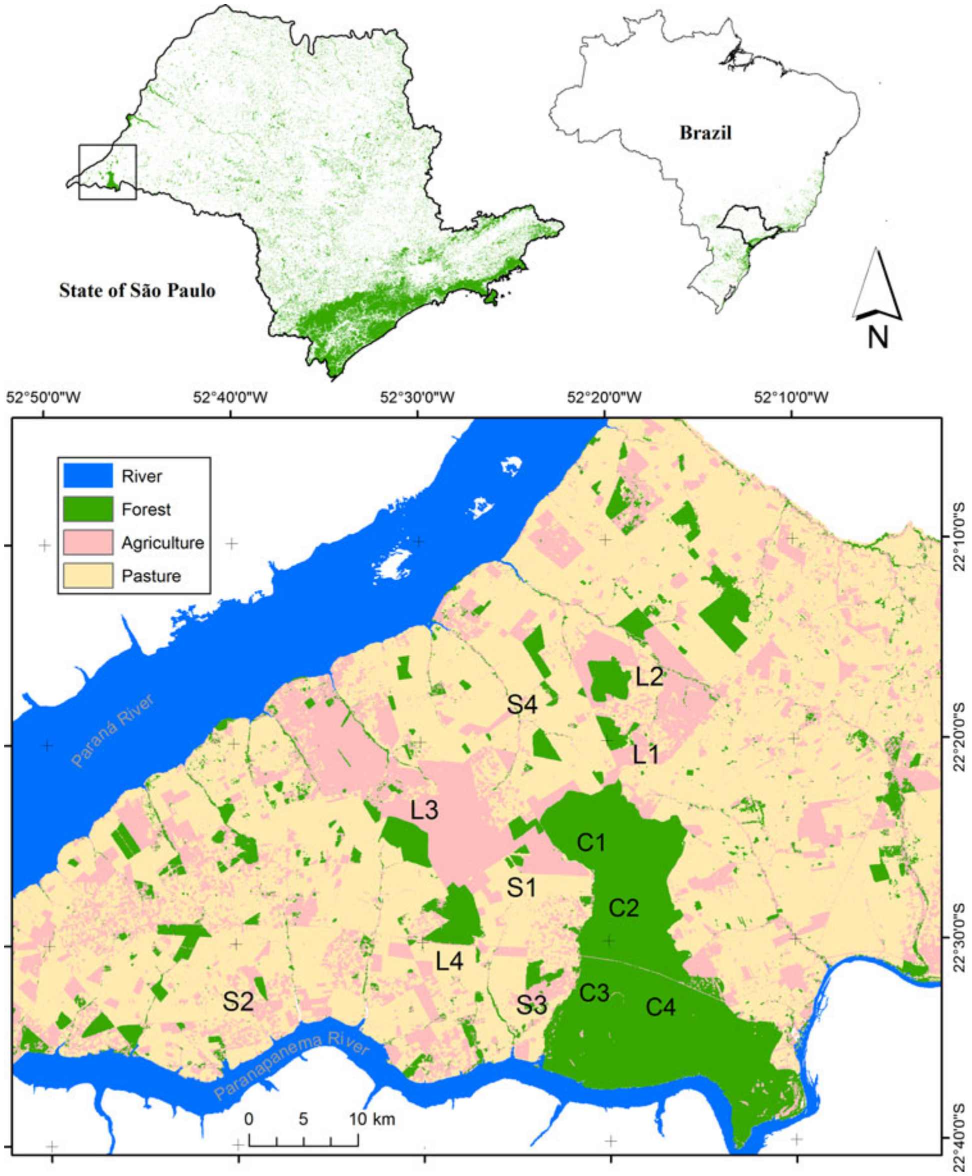

Figure 1. Study area of Pontal do Paranapanema Region, SP, Brazil. Small patches: S1, S2, S3, S4; large patches: L1, L2, L3, L4; Control areas inside the Morro do Diabo State Park: C1, C2, C3, C4.

Tick identification was performed as described in Ogrzewalska et al. (2009a), who reported raw data on the ticks collected in the present study. In addition, the rickettsial infection of these ticks was previously reported in Ogrzewalska et al.(2009b), who applied polymerase chain reaction (PCR) and DNA sequencing for detection and identification of rickettsiae infecting the ticks (detailed techniques were described by the authors). Herein, we analyzed both these tick and rickettsial data in relation to forest fragmentation.

Data Analyses

For each sample site, we calculated the following parameters: bird richness, Shannon diversity index, bird abundance (number of individuals per 100 net-hours), and the percentage of bird individuals infested with ticks (number of infested birds/number of examined birds × 100, for each bird species).

To test for the effect of habitat reduction on bird species, we used the Kruskal–Wallis test with the a posteriori Dunn's Test. For birds that occurred in more than seven sampled areas, if the abundance was higher in control and large fragments in relation to small fragments, we considered the species affected by fragmentation. If the abundance was constant between different size categories, we considered the species not affected. If the abundance was higher in small fragments, we considered the species to have benefited. For species that occurred in four to seven sampled areas, categorization was made based on presence/

absence. Species that occurred only in control and large fragments were considered affected. If species were present in all patch size classes, they were considered not affected. When species were present only in small areas, they were considered to have benefited. For species that occurred in less than four sampled areas, or if it was not possible to determine their response through the criteria mentioned above, the effect of fragmentation was determined based on data from previous studies in the same region (Uezu and Metzger 2011).

To compare the richness and diversity of birds and the prevalence of ticks on birds between the size categories, we again used the Kruskal–Wallis test with the a posteriori Dunn's test.

The influence of the richness and diversity of birds, and abundance of the most common birds infested by ticks (*Conopophaga lineata* and *Thamnophilus pelzelni*) on the prevalence of ticks in each studied area was verified through simple linear regressions.

To test for the influence of forest fragment size on the abundance of ticks in the environment, we used the same procedures described above for birds. However, for these regression analyses, values from control areas were averaged and represented by a single point as the control areas were located inside the same continuous forest. Tick infection rates by *Rickettsia* were compared between the forest fragments using a Chi-square test.

RESULTS

We captured 1,725 birds representing 80 species from 24 families within 10 orders, excluding recaptures from the same day. The most abundant species was *T. pelzeni* (317 individuals), followed by *C. lineata* (150 individuals) and *Arremon flavirostris* (147 individuals) (Table 1).

Table 1. The number of individuals caught of the 20 most abundant bird species caught by mist nets from December 2004 to December 2006

Species	Frequency
Thamnophilus pelzeni	317
Conopophaga lineata	150
Arremon flavirostris	147
Pipra fasciicauda	145
Platyrinchus mystaceus	94
Cnemotriccus bimaculatus	78
Baryphthengus ruficapillus	76
Basileuterus culicivorus	69
Sittasomus griseicapillus	66
Basileuterus flaveolus	64
Corythopis delalandi	64
Dysithamnus mentalis	43
Leptopogon amaurocephalus	27
Leptotila verreauxi	21
Pyriglena leucoptera	21
Malacoptila striata	19
Picumnus albosquamatus	19
Thalurania glaucopis	18
Thamnophilus caerulescens	18
Turdus amaurochalinus	18

Bird species showed different responses to fragmentation, which we categorized into three classes: (1) sensitive species that were present only in the control areas and/or large fragments; (2) unaffected species whose abundance did not change between sampled areas; and (3) benefited species that increased their abundance in smaller fragments. Species such as *Corythopis delalandi, Malacoptila striata, Schiffornis virescens, Xenops rutilans*, *Nonnula rubecula, Hemitriccus orbitatus*, and *Chiroxiphia caudata* were present only in control areas and/or in large fragments and thus were considered sensitive to fragmentation. Similarly, species such as *Baryphthengus ruficapillus, Pipra fasciicauda,* and *Platyrinchus mystaceus* were significantly ($P < 0.05$) more abundant in control than in smaller fragments. Other species, such as *T. pelzelni, C. lineata, A. flavirostris, Basileuterus culicivorus*, *Basileuterus flaveolus, Sittasomus griseicapillus, Dysithamnus mentalis, Leptotila verreauxi,* and *Pyriglena leucoptera* maintained the same abundance between sampled areas, being not significantly affected by fragmentation ($P > 0.05$) (Fig. 2). Conversely, species such as *Cnemotriccus fuscatus*, *Hemitriccus margaritaceiventer*, *Formicivora rufa* and *Cyanocorax chrysops* benefited from a fragmented landscape, having significantly higher abundances in smaller patches ($P < 0.05$).

Bird diversity was influenced by fragment size, being lower in smaller fragments [$H_{(12.2)} = 6.27$; $P = 0.043$] (Fig. 3). Similarly, a tendency for lower richness in smaller fragments was also observed (Fig. 3), but the P value was slightly higher than assumed criteria [$H_{(12.2)} = 5.88$; $P = 0.053$].

A total of 223 (13%) birds were found infested by 1, 800 larvae and 539 nymphs of the genus *Amblyomma*. Identification of these ticks were as follows: *Amblyomma nodosum* (58 larvae, 394 nymphs), *Amblyomma longirostre*

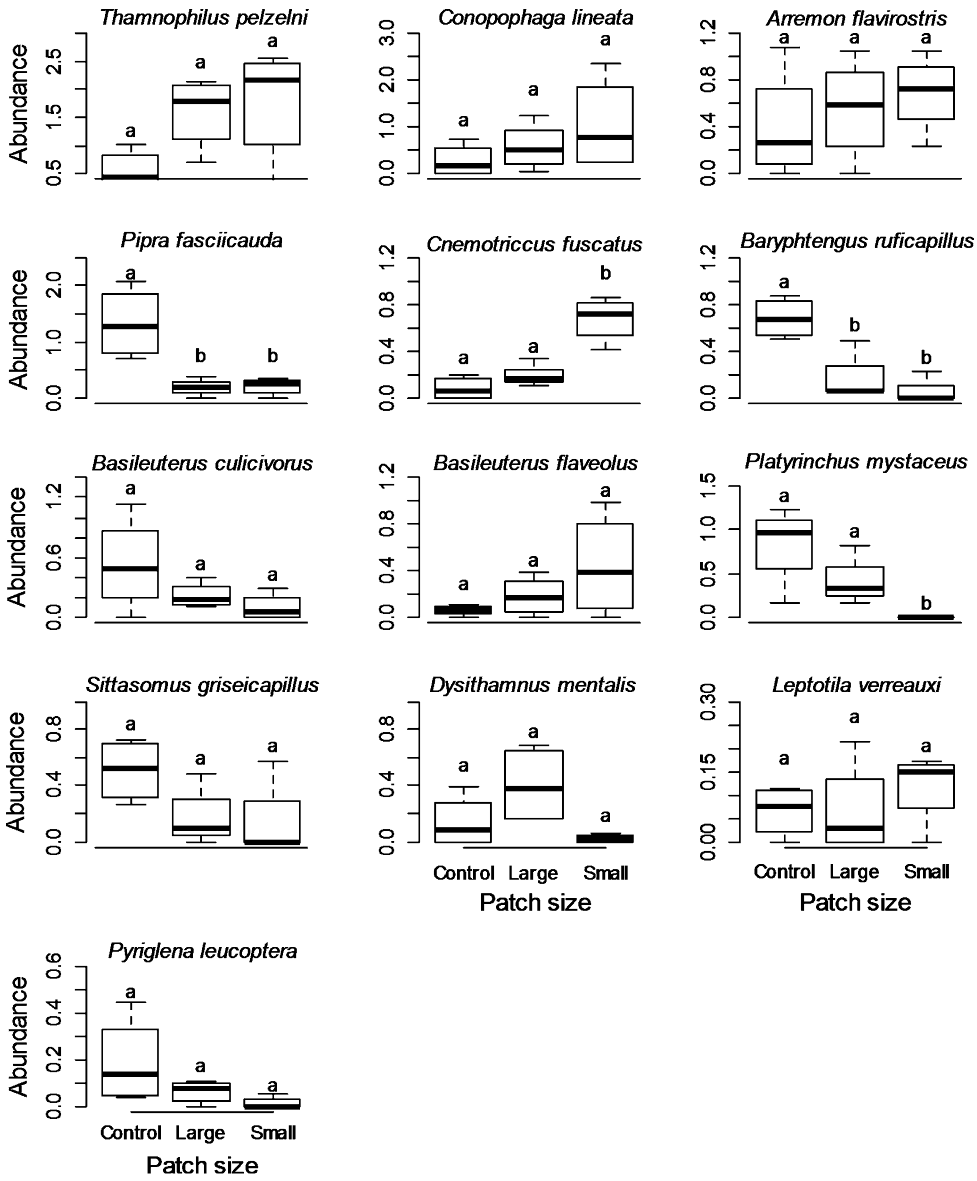

Figure 2. Variability by patch size in the abundance of the 13 most common birds caught by mist nets. The limits of the *boxes* indicate the first and third quartile and *lines* indicate the minimum and maximum abundance. Results with *different letters* indicate statistically significant differences (i.e., $P < 0.05$).

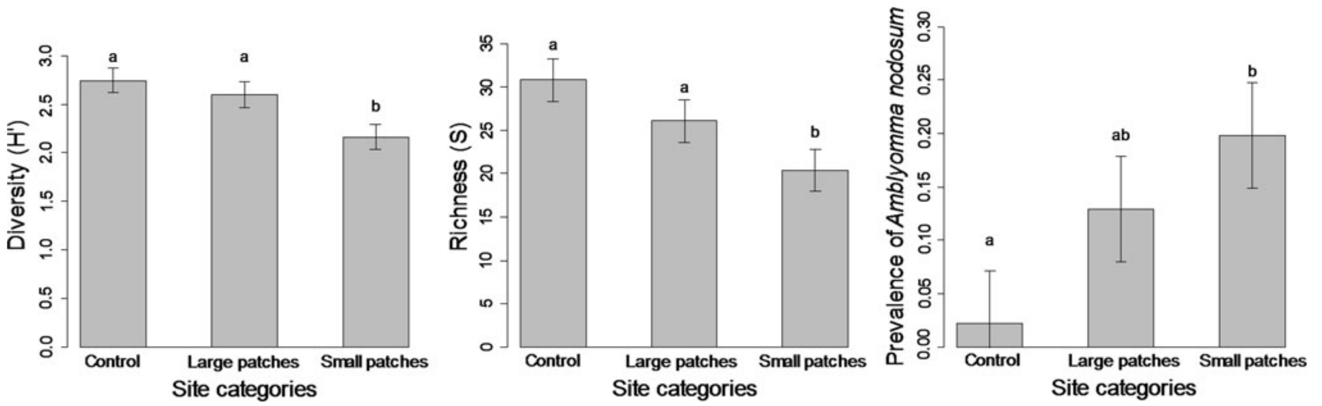

Figure 3. Variation in bird diversity, richness and in the prevalence of *Amblyiomma nodosum* among sampled sites in Pontal do Paranapanema region. *Lines* indicate standard errors. Results with *different letters* indicate statistically significant differences (i.e., $P < 0.05$).

(4 larvae, 3 nymphs), *A. cajennense* (12 nymphs), *Amblyomma calcaratum* (2 nymphs), *Amblyomma coelebs* (11 larvae, 12 nymphs), *Amblyomma ovale* (5 larvae, 5 nymph), and *Amblyomma naponense* (1 nymph). A total of 1,722 larvae and 110 nymphs collected from birds could not be identified to species and were regarded as *Amblyomma* sp. Detailed data are presented elsewhere (Ogrzewalska et al. 2009a).

We found an inverse relationship between the prevalence of ticks on birds and the diversity and richness of birds ($R^2 = 0.51$; $P = 0.032$ and $R^2 = 0.44$; $P = 0.052$, respectively). When diversity and richness were higher, the prevalence of ticks was lower (Fig. 4).

We also found a positive relationship between the prevalence of ticks on birds and the abundance of *C. lineata* ($R^2 = 0.39$; $P = 0.03$; Fig. 4), although this pattern was not present for *T. pelzelni* ($R^2 = 0.01$; $P = 0.74$).

The prevalence of the most common tick to parasitize birds, *A. nodosum*, was influenced by the size of forest fragments, being higher in small fragments [$H_{(12.2)} = 6.64$; $P = 0.036$; Fig. 3]. We observed that *A. cajennense* and *A. longirostre* occurred in all three classes of patch size, while *A. coelebs*, *A. calcaratum*, and *A. naponense* occurred only in control and/or in large fragments, and *A. ovale* was more frequent in small fragments (Table 2). Since the occurrence of other tick species on birds was verified in only a few forest fragments, the effect of fragmentation on these ticks could not be tested statistically.

Among ticks collected from the environment *A. cajennense* was the most abundant, represented by 227 adults, 1,992 nymphs, and 31 larval clusters, representing 65.5% of all field-collected ticks. Other free-living species, *A. coelebs, A. naponense, A. brasiliense,* and *Haemaphysalis juxtakochi*, occurred only occasionally, representing 4.2% of all identified ticks. The remaining 30.3% of the free-living ticks were not identified to species and were regarded as *Amblyomma* sp. *A. cajennense* occurred in all sampled areas with the exception of one small fragment (P2). However, its abundance varied significantly, being higher in control and large fragments ($R^2 = 0.70$; $P < 0.01$; Fig. 4). *A. coelebs, A. brasilense*, and *A. naponense* occurred more frequently in control and large fragments, and sporadically in small fragments, while *H. juxtakochi* occurred in small numbers in fragments of different size categories (Table 2).

Among 174 nymphs of *A. nodosum* that were tested for rickettsial infection, 41 (23.6%) were found infected with *Rickettsia parkeri* strain NOD and 10 (5.7%) with *Rickettsia bellii*. Detailed data about molecular characterization and isolation of these bacteria from the *A. nodosum* ticks have

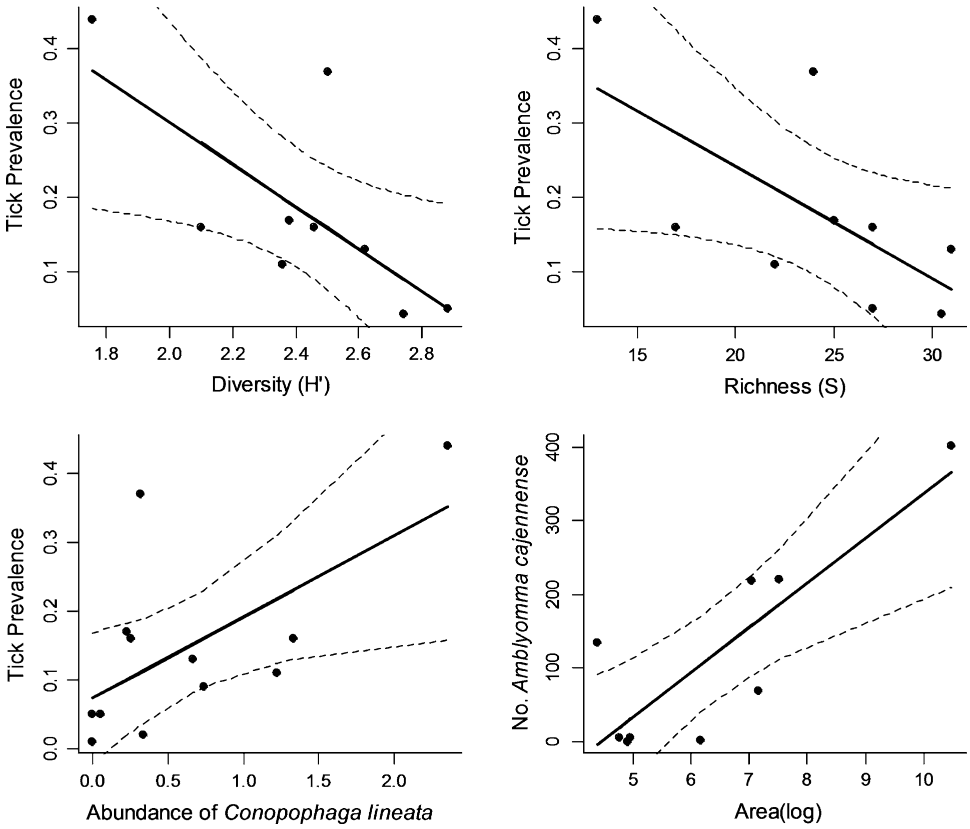

Figure 4. Variation in tick prevalence on birds according to bird species diversity and richness, abundance of *Conopophaga lineata* and abundance of *Amblyomma cajennense* among sampled sites in Pontal do Paranapanema region. *Dashed lines* show 95% confidence intervals.

Table 2. Number of ticks collected on birds or from the environment in three different patch size areas (Control, Large, and Small) in the Pontal do Paranapanema region, Brazil

Ticks species	Areas					
	Control		Large		Small	
	Bird	Environment	Bird	Environment	Bird	Environment
A. nodosum	25N	–	91N	–	278N, 58L	–
A. coelebs	2N, 11L	5A, 28N, 4C	10N	2A, 14N, 2C	–	1A, 5N
A. longirostre	1L	–	3L	–	3N	–
A. cajennense	8N	131A, 1443N, 24C	2N	78A, 426N, 6C	2N	18A, 123N, 1C
A. ovale	–	–	2N	–	3N, 5L	–
A. naponense	–	1A, 25N, 2C	1 N	4A, 7N	–	2A, 2N
A. calcaratum	2N	–	–	–	–	–
A. brasilense	–	4A, 13N, 4C	–	2A, 7N	–	3A, 2N
Amblyomma spp	9 N	490N, 26C	89N, 1661L	299N, 5C	12 N, 61L	221N, 1C
H. juxtakochi	–	2N	–	1 N	–	1 N
Total	46N, 12L	141A, 2001N, 60C	195N, 1664L	86A, 754N, 13C	298N, 124L	24A, 354N, 2C

A adult ticks, *N* nymphs, *L* larvae, *C* larval clusters, – no tick.

Table 3. Number of *Amblyomma nodosum* nymphs infected by *Rickettsia parkeri* or *Rickettsia bellii* according to the fragment size where the ticks were collected on birds in the Pontal do Paranapanema region, Brazil

Areas	No. of infected nymphs/No. tested (prevalence)	
	Rickettsia parkeri	*Rickettsia bellii*
Control	2/6 (33.3)	0/6 (–)
Large	6/38 (15.8)	3/38 (7.9)
Small	33/130 (25.4)	7/130 (5.4)
Total	41/174 (23.6)	10/174 (5.7)

been reported elsewhere (Ogrzewalska et al. 2009b). *R. parkeri* was found in 33.3, 15.8, and 25.8% of the nymphs from control, large, and small fragments, respectively. These infection rates were statistically similar ($\chi^2 = 1.83$; DF $= 2$; $P = 0.400$). *R. bellii* was found in 7.9 and 5.5% of the nymphs from the large and small fragments, respectively ($\chi^2 = 0.33$; DF $= 1$; $P = 0.565$). It was not found in the control area (Table 3).

DISCUSSION

During the last 50 years, the study region of Pontal do Paranapanema has suffered dramatic deforestation. The landscape is now composed predominantly of pastures and sugar cane plantations, with less than 20% of the original forest cover left (Leite 1998). Such changes cause drastic habitat fragmentation, leading to isolation and a decrease of animal populations (Fahrig 2003). In other regions, such ecological modifications are known to have altered the relationships between vertebrate hosts and their parasites, affecting their behaviors, movements, densities, and consequently influencing the transmission of vector-borne pathogens (Ostfeld and Keesing 2000).

In the present study, we verified that the diversity and richness of wild bird species were lower in smaller forest fragments, a finding that is in concordance with a previous study conducted in the same region (Uezu and Metzger 2011) and with other studies of avifauna (e.g., Willis 1979; Ribon et al. 2003; Uezu et al. 2005). Some more vulnerable bird species survive only in larger forest remnants, with the small and isolated fragments having become too small to sustain local populations and the birds being unable to cross the non-habitat portion of the landscape to maintain dispersal with other populations, leading to local extinction (Fahrig 2003).

Ticks are also affected by forest fragmentation. Considering the most abundant tick species found on birds (*A. nodosum*) or in the environment (*A. cajennense*), our results showed that the abundance of *A. nodosum* on birds was significantly higher in the fragmented landscape, while the abundance of *A. cajennense* on the vegetation was significantly lower. Previous studies with other ticks species in other biomes showed that the abundance of the main mammalian hosts has a direct effect on tick abundance. In other words, the higher the host abundance, the higher the chances that ticks will find the hosts to complete their life cycle, increasing the tick population (Randolph 2004; Deblinger et al. 1993; Ostfeld and Keesing 2000; Allan et al. 2003; Rizzoli et al. 2009).

Amblyomma nodosum is a poorly studied Neotropical tick, for which the adult stage feeds chiefly on the anteaters *Tamandua tetradactyla* and *Myrmecophaga tridactyla*, with understory Passeriformes birds serving as hosts for the immature stages (Labruna et al. 2007; Ogrzewalska et al. 2009a). In the present study, the birds *T. pelzelni* and *C. lineata* were the most frequent hosts of immature *A. nodosum* in all fragments sampled (small, large, and control areas). Although the overall abundance of these two bird species were not significantly different between control, large, and small areas (Fig. 2), a total of 147 *T. pelzelni* and 84 *C. lineata* were captured in small fragments during the study, in contrast to only 43 *T. pelzelni* and 19 *C. lineata* captured in the control areas. Thus, it is possible that higher host availability in the small fragments could have accounted for the higher abundance and prevalence of *A. nodosum* infestations on birds in these small forest areas. The prevalence of ticks on birds is also significantly correlated with the abundance of *C. lineata*, supporting this conjecture.

The two main hosts of adult *A. nodosum* ticks, *M. tridactyla* and *T. tetradactyla*, were known to occur in our study region (Lima 2009). In this case, different abundance of these mammals could also have affected our results. It is possible that abundance of anteaters was higher in small fragments, due to higher food availability (ants and termites) or because of the lack of predators like ocelots and jaguars (Redford and Eisenberg 1989), which are the first to disappear in fragmented landscapes (Noss et al. 2005). However, even though *M. tridactyla* and *T. tetradactyla* are found in all Brazilian biomes, where they are able to use many types of vegetation from tropical rainforest to savannahs, we are aware of no studies concerning their abundance. Therefore, it is not possible to confirm if their populations were influenced by habitat fragmentation or if *A. nodosum* was affected by anteater abundance.

Another factor that could have affected the distribution of ticks is the microclimate of the fragments. The ticks found in the present study take only one blood meal per life stage, as larva, as nymph, and as adult. Each feeding stage takes no more than a few days on the host, when the ticks drop off to develop to the next stage, which might take weeks to months between successive stages. Thus, despite being parasites, these ticks spend most of their life cycle in the environment, where they also take time to search for their hosts and are strongly influenced by environmental conditions (Oliver 1989; Randolph 2004). Successful development of free-living stages is affected primarily by temperature and humidity, which have been shown to affect tick abundance (Randolph 2004). Although there are no data on optimal microclimatic requirements for *A. nodosum*, it is suspected that this species, which commonly parasitizes anteaters in the Cerrado biome (a Brazilian savannah less humid than rainforest), finds more optimal conditions in smaller and degraded forest patches. Such smaller fragments are strongly influenced by their surroundings and, because of a less dense canopy, the temperature should be higher and the humidity lower when compared with the conditions of the original Atlantic Forest.

The most common tick found in the environment, *A. cajennense*, feeds on a wide range of animals, although larger mammals such as tapirs (*Tapirus terrestris*) and peccaries (*Tayassu* spp.) are preferable for adult ticks (Aragão 1936; Estrada-Pena et al. 2004). The highest abundance of this tick species was observed in the control areas, while the species seems to vanish from small and isolated fragments, suggesting that its principal wild hosts (tapirs and peccaries) have already disappeared from these small fragments. This disappearance was recently confirmed in a field study on peccaries in the same forest fragments of the present study (Nava 2009). A similar tendency was observed for other tick species found in the environment, such as *A. coelebs* that parasitizes mainly tapirs, and *A. naponense* and *A. brasiliense* that parasitize chiefly peccaries (Labruna et al. 2005; Szabo et al. 2009). These results confirm that these medium to large-sized mammals are also disappearing from small forest fragments, perhaps leading to the local co-extinction of their ticks. In this case, our results confirm that questing ticks can be used as bio-indicators of wild fauna (Labruna and Guglielmone 2009).

In the studied region, 5.7 and 23.6% of *A. nodosum* nymphs collected on birds were infected with *R. parkeri* and *R. bellii*, respectively (Ogrzewalska et al. 2009b). *R. bellii* is the most commonly found rickettsia in Neotropical ticks, and there is no evidence that it is pathogenic for humans or animals (Labruna 2009). On the other hand, *R. parkeri* is responsible for spotted fever cases in humans in the United States (Paddock et al. 2004), Uruguay (Conti-Díaz et al. 2009), and Brazil (Spolidorio et al. 2010). In Brazil, different strains of *R. parkeri* have been reported in ticks, including the strain NOD, also detected in *A. nodosum* ticks from our study region (Ogrzewalska et al. 2009b).

CONCLUSIONS

The proportion of *Rickettsia*-infected ticks found in our study was not exceptionally high. However, forest fragmentation showed a significant effect on tick abundance and on bird richness and abundance. While there did not appear to be a significant change in the actual rates of Rickettsia infection in *A. nodosum* ticks, the greater absolute abundance of ticks likely increases the risk of Rickettsia contagion. These results probably occurred because regardless of the fragment size, the *A. nodosum* population tended to feed mostly on the same host species. Additionally, it is possible that the main perpetuation route of *R. parkeri* strain NOD among *A. nodosum* ticks is through vertical transmission, as reported for other rickettsial strains in other tick species (Parola et al. 2005). Thus, horizontal transmission via vertebrate hosts could be of little importance for this rickettsia. We conclude that the higher burdens of immature *A. nodosum* ticks on wild birds in small forest remnants, and the tendency of other ticks typical for ungulates (especially *A. cajennense*) to disappear from these small fragments, are important effects of habitat fragmentation, showing the imbalance between the environment–host–parasite relationship.

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