Pterygosomatidae and Trombiculidae mites infesting *Tropidurus hispidus* (Spix, 1825) (Tropiduridae) lizards in northeastern Brazil

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Abstract

Parasitism of the lizard *Tropidurus hispidus* by *Geckobiella* sp. and by larvae of *Eutrombicula alfreddugesi* was examined in a mountainous area in Chapada do Araripe (07° 16' S and 39° 26' W), southern Ceará State, Brazil. Of the 56 lizards collected (26 females, 27 males, and 3 juveniles), 40 (total prevalence of 71.42%) were infested by mites. Mite-pockets were the sites most heavily infested by *E. alfreddugesi* larvae, while *Geckobiella* sp. was found uniformly distributed under scales over the host's entire body. The female specimens of *T. hispidus* parasitised by *E. alfreddugesi* had an average infestation rate of 8.57 ± 3.62, 1-27, while the males had an average infestation rate of 11.90 ± 2.63, 1-25. The female specimens parasitised by *Geckobiella* sp. had an average infestation rate of 5.91 ± 2.28, 1-25, while the males had an average infestation rate of 5.43 ± 1.52, 1-23. Seven specimens were also infested by eggs and immature forms of unidentified mites (average 2.28 ± 0.89, 1-7). There were no significant differences between the total prevalence of mites on adult male (70.4%) and adult female (65.4%) lizards. The body sizes of the hosts did not influence their infestation rates. The average infestation intensity by *E. alfreddugesi* (10.2 ± 8.7) was significantly greater than the average infestation intensity by *Geckobiella* sp. (5.9 ± 6.8). *T. hispidus* is the new host record to *Geckobiella* mites.

Keywords: mites, *Eutrombicula*, *Geckobiella*, lizards, *Tropidurus.*

Ácaros Pterygosomatidae e Trombiculidae infestando lagartos *Tropidurus hispidus* (Spix, 1825) (Tropiduridae) no nordeste do Brasil

Resumo

No presente estudo foi analisado o parasitismo do lagarto *Tropidurus hispidus* pelos ácaros *Geckobiella* sp. e larvas de *Eutrombicula alfreddugesi* em uma área na Chapada do Araripe (07° 16' S e 39° 26' W), região sul do Estado do Ceará, Brasil. Dos 56 lagartos coletados (26 fêmeas, 27 machos, e 3 juvenis), 40 (prevalência total de 71,42%) estavam infestados por ácaros. Entre os *sites* de infestação, as bolsas de ácaros foram os mais infestados pelas larvas de *E. alfreddugesi*, enquanto *Geckobiella* sp. foi encontrado distribuído uniformemente sob as escamas por todo o corpo dos hospedeiros. Os espécimes fêmeas de *T. hispidus* parasitados por *E. alfreddugesi* tinham uma infestação média de 8,57 ± 3,63, 1-27, enquanto os machos tinham uma média de infestação de 11,90 ± 2,63, 1-25. Os espécimes fêmeas parasitados por *Geckobiella* sp. tinham uma infestação média de 5,91 ± 2,28, 1-25, enquanto que os machos tinham uma infestação média de 5,43 ± 1,52, 1-23. Sete espécimes estavam também infestados por ovos e formas imaturas de ácaros não identificados (infestação média de 2,28 ± 0,89, 1-7). Não houve diferenças significativas entre a prevalência total de ácaros em machos (70,4%) e fêmeas (65,4%) adultas. As médias dos tamanhos corporais não influenciaram as taxas de infestação. A intensidade de infestação média por *E. alfreddugesi* (10,2 ± 8,7) foi significativamente maior do que a encontrada para *Geckobiella* sp. (5,9 ± 6,8). *T. hispidus* constitui um novo registro de hospedeiro para ácaros do gênero *Geckobiella*.

Palavras-chave: ácaros, *Eutrombicula*, *Geckobiella*, lagartos, *Tropidurus.*

1. Introduction

Lizards and parasitic mites demonstrate ancient relationships, so much so that some lizards have developed skin folds (through independent phylogenetic events) that form structures known as "mite-pockets" in different regions of their bodies where mites tend to aggregate (Rodrigues, 1987; Bauer, 1990, 1993). Arnold (1986) argued that the development of these "mite pockets" acts as an adaptation to limit the distribution of trombiculidae ectoparasites on their bodies and thus reduce their damage. However, scansorial mites such as *Geckobiella* spp. (as well as other pterygosomatids) are not usually found in mite-pockets (Bauer, 1990, 1993; Bertrand and Modry, 2004).

Among the parasitic mites found on lizards, the family Pterygosomatidae comprises nine genera, only one of which has to date been found on lizards (*Pimeliaphilus*) (Bertrand and Modry, 2004). *Geckobiella texana* (Banks, 1904) has herpetological importance among the pterygosomatid ectoparasites since it is a vector of the protozoarian *Schellackia occidentalis* Bonorris and Ball, 1955, found in the intestines and blood tissue of lizards (Bonorris and Ball, 1955). Trombiculidae represents one of the most widely distributed families in the Neotropical Region (Zippel et al., 1996, Daniel and Stekolnikov, 2004; Klukoswski, 2004), presenting interactions mainly of *Eutrombicula alfreddugesi* (Oudemans, 1910) with a few other lizard species. In Brazil, the larvae of this chigger mite have been reported as ectoparasites on lizards in 'restinga' (sandy coastal) vegetation formations (Cunha-Barros and Rocha, 1995, 2000; Cunha-Barros et al., 2003), on tropidurid lizards from 'cerrado' (savanna) vegetation regions (Carvalho et al., 2006), and in the ecotone between the 'caatinga' (dry thorny shrub/stunted trees) and 'campos rupestres' (higher altitude rocky field) habitats (Rocha et al., 2008). Chiggers (the larvae of the Trombiculidae) are important pests on some groups of reptiles and cause dermatitis, blood loss, and are vectors of infectious diseases (Frye, 1991).

Price (1980) argued that vacant niches still exist for parasites and that niches of coexisting parasites are mostly non-overlapping. For an arthropod ectoparasite, for example, the host surface constitutes its habitat, and its mode of occupation will depend on intrinsic and extrinsic factors acting at that environment scale (Marshall, 1981; Bittencourt and Rocha, 2002). In some cases, different ectoparasite species can share different portions of the surface body of a host, with parasite species presenting different niche width and overlap in microhabitats of host body surface (Bittencourt and Rocha, 2003).

We investigated the infestation patterns (prevalence and intensity; sensu Bush et al., 1997) of the mites *Geckobiella* sp. and *Eutrombicula alfreddugesi* on the lizard *Tropidurus hispidus* in a mountainous region of Chapada do Araripe, Brazil, to evaluate which microhabitats were occupied and to what extent are there spatial niche overlaps between these mite species. Additionally we evaluated the relationship between i) host body size and ii) host sex with infestation rates.

2. Material and Methods

Field work was carried out on the lower slopes of the Chapada do Araripe Mountains (07° 16' S and 39° 26' W) within the Chapada do Araripe Environmental Protection Area, in the municipality of Crato, Ceará State, Brazil. The regional vegetation there is a mosaic of palm trees and montane and secondary forests. The area has experienced anthropogenic alterations resulting from agricultural use, the harvesting of natural products, and land development. The regional climate is warm, semi-arid tropical. Mean annual temperatures range from 24° to 26° C. The rainy season extends from January to May, and the mean annual rainfall is 1090 mm (IPECE, 2008).

Lizards were captured by hand using rubber slings or nooses. Immediately after capture, each lizard was transferred to a plastic sack containing cotton soaked in ether that anaesthetized and euthanized them. Their snout-vent lengths (SVL) were measured (to the nearest 0.05 mm) using calipers. The lizards were subsequently fixed in 10% formalin and stored in 70% alcohol.

All of the external surfaces of the lizards' bodies were carefully searched for mites using a stereomicroscope. The number and the position of the mites on the lizard bodies (mite microhabitats on host body) were recorded in order to identify and to map the specific occupied sites. The following sites were designated in this mapping analysis: Forearm (Fa), Doral Face of Head (DFH), Ventral Face of Head (VFH), Thigh (Th), Dorsal (Do), Ventral (Ve), Lateral neck pocket (Lnp), Auxiliary pocket (Pax), Inguinal Mite Pocket (Pin), Arm (Ar), Inside Elbow (IE), Inside knee (IK), Pre-Femoral Region (PFR), Leg (Lg), Dorsal Face of the Tail (DFt), Ventral Face of the Tail (VFt) (Figure 1).

The mites were collected using forceps and fine-bristle brushes and mounted on permanent slides in Hoyer medium for subsequent identification.

Statistical tests were performed utilising: the Z test for proportions (Zar, 1999) to evaluate if there were significant differences in the overall prevalence of mites on male and female lizard hosts. We also used Student *t*-test (Zar, 1999) to evaluate if there were significant differences in the mean infestation intensity among host sexes, and between *E. alfreddugesi* and *Geckobiella* sp.

Principal component analysis (PCA) (Jongman et al., 1995) was performed to evaluate the relationships between the mite species and their infestation sites, using the MVSP 3.1 software program (Kovach, 1999).

We used Pianka's measure of niche overlap (Pianka, 1973) to evaluate the overlap in spatial niches between the two mite species (Equation 1):

$$O_{12} = O_{21} = \frac{\sum_{i=1}^{n} P2iP1i}{\sqrt{\sum_{i=1}^{n}\left(P2i^2\right)\left(P1i^2\right)}} \quad (1)$$

where P2i and P1i are the rate of use of microhabitat type i by mite species 1 and 2 respectively. We compared the observed overlap values against a null model (1000 interactions) generated by the R3 randomization algorithm (Lawlor, 1980) using ECOSIM 7.0 software (Gotelli and Entsminger, 2001).

3. Results

A total of 56 specimens of *T. hispidus* were collected, including 26 females (SVL 77.84 ± 2.33, 55-95 mm), 27 males (SVL 95.29 ± 2.57, 70-113 mm), and 3 juveniles (SVL 49.66 ± 0.33, 49-50 mm).

Forty lizards were infested by at least one mite species; overall prevalence was 71.4%.

Two mite species, *E. alfreddugesi* (Trombiculidadae) and *Geckobiella* sp. (Pterygosomatidae) were found.

Host females parasitised by *E. alfreddugesi* had a mean infestation intensity of 8.57 ± 3.62 (range 1-27). Infested host males had a mean infestation intensity of 11.90 ± 2.63 (range 1-25). Female lizards infested by *Geckobiella* sp. had a mean intensity of infestation of 5.91 ± 2.28, 1-25, whereas males had a mean infestation intensity of 5.43 ± 1.52, 1-23. The three juvenile specimens were parasitised exclusively by *E. alfreddugesi*, and their mean infestation intensity was 8.0 ± 4.72, 1-17. Seven adult lizards were infested by eggs and immature forms of unidentified mites (mean 2.28 ± 0.89, 1-7).

There was no significant difference between the overall infection rates of adult male (70.4%) and adult female (65.4%) lizards (Z-test: zc = 1.44; p = 0.925).

The overall mean infestation intensity of adult male lizards (12.3 ± 8.4) was not significantly different from the overall mean infestation intensity of females (7.9 ± 9.2) (*t*-test: t = -1.62; g.l. = 35; $p > 0.05$).

The mean intensity rate of infestation by *E. alfreddugesi* (10.2 ± 8.7) was significantly higher than that by *Geckobiella* sp. (5.9 ± 6.8) (*t*-test: t = -1.94; g.l. = 47; $p < 0.05$).

The first two axes of the PCA explained 99.9% of the observed variance (Table 1). Axis 1 ordered the mite species principally in regards to their abundance in mite-pockets (Lnp) and in auxiliary-pockets (Pax) in the direction of positive values (Table 2, Figure 2). Axis 2 ordered the mite species principally in relation to their abundance on abdomen (Ab), thigh (Th), and leg (Pr) sites, with positive values (Table 2, Figure 2).

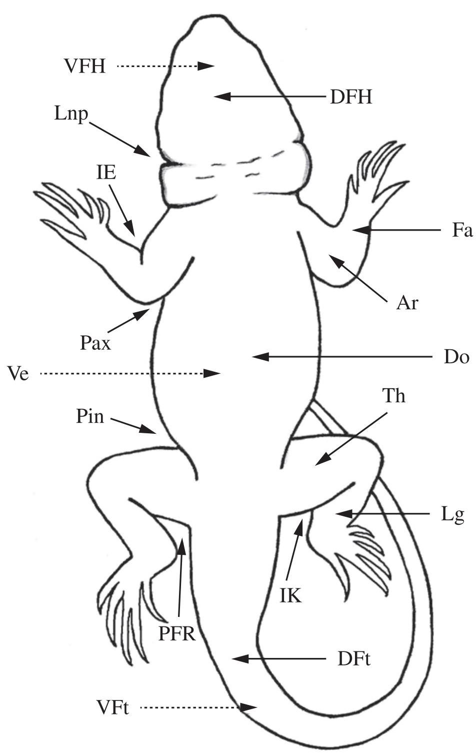

Figure 1. Main infestation microhabitats of *Eutrombicula alfreddugesi* and *Geckobiella* sp. on the body of the lizard *Tropidurus hispidus*: LEGEND: ventral side of the head = VFH; forearm = Fa; axially-pocket = Pax; mite-pocket = Lnp; inguinal-pocket = Pin; arm = Ar; Thigh = Th; forearm/ inner elbow = IE; leg fold/thigh = IK; dorsal = Do; Dorsal side of the head = DFH; dorsal side of the tail = DFt; ventral side of the tail = VFt; leg = Lg; pre-femoral region = PFR; Ventral region = VE.

Table 1. Autovalues and the percentage of variance that was explained by the two principal components (axes 1 and 2) of the Principal Component Analysis (PCA) for the abundance of *E. alfredugesi*, *Geckobiella* sp., and immature mite forms at diverse infection sites on the host *Tropidurus hispidus*.

	Axis 1	**Axis 2**
Autovalues	13055.3	1584.7
Percentages	89.1	10.8
Accumulated percentages	89.1	99.9

Table 2. Autovectors of the two principal components (axes 1 and 2) of the Principal Component Analysis (PCA) of the abundance of *E. alfredugesi*, *Geckobiella* sp., and immature mite forms at diverse infection sites on the host *Tropidurus his*. The most important variables on axes 1 and 2 are indicated in bold type. LEGEND: ventral side of the head = VFH; forearm = Fa; axially-pocket = Pax; mite-pocket = Lnp; inguinal-pocket = Pin; arm = Ar; Thigh = Th; forearm/ inner elbow = IE; leg fold/thigh = IK; dorsal = Do; Doral side of the head = DFH; dorsal face of the tail = DFt; ventral side of the tail = VFt; leg = Lg; pre-femoral region = PFR; Ventral region = VE.

Site	**Axis 1**	**Axis 2**
VFH	0.00	0.02
Ab	0.00	**0.11**
Pax	**0.41**	0.07
Lnp	**0.91**	-0.07
Pin	0.00	**0.11**
Br	0.00	**0.16**
Th	0.01	**0.44**
IE	0.00	**0.12**
IK	0.01	**0.25**
Do	0.01	**0.29**
DFH	0.00	0.02
DFt	0.00	0.02
VFt	0.00	0.09
Lg	0.01	**0.30**
PFR	0.01	0.03
VE	0.03	**0.70**

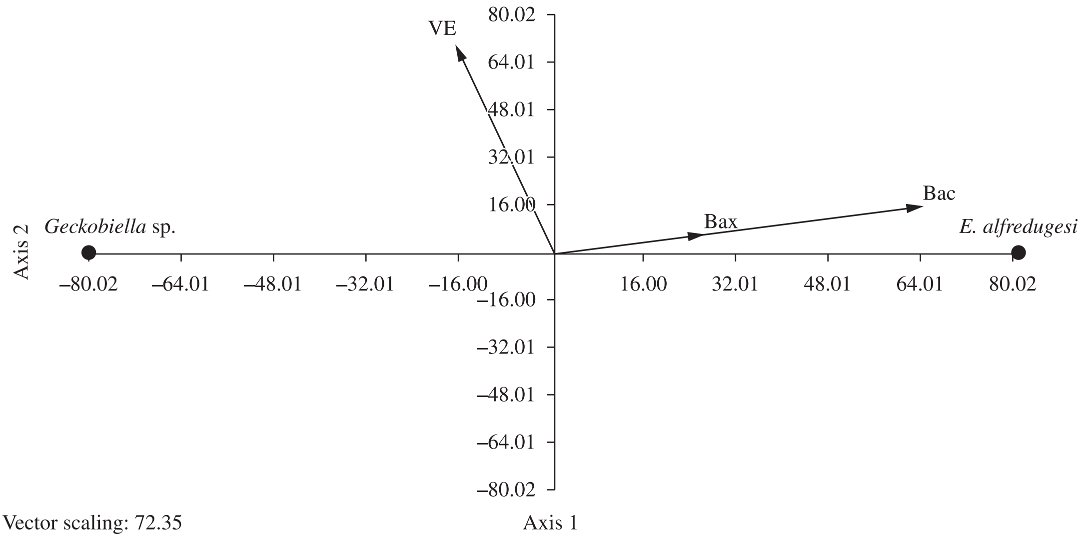

Figure 2. Ordination diagram ("biplot") of the abundances of *E. alfredugesi*, *Geckobiella* sp., and immature mite forms along the 1° and 2° axes of the Principal Component Analyses (PCA) in relation to their various infection sites on *T. hispidus*.

The spatial niche overlap between *E. alfreddugesi* and *Geckobiella* sp. was $O_{12} = O_{21} = 0.197$. This value did not differ significantly from that generated by null models (RA3; 1000 iterations; $O_{12} = O_{21} = 0.248$; p = 0.50).

4. Discussion

The ectoparasite species *E. alfreddugesi* has been found in all studies of mite infestations on lizards in Brazil, and their infestation rates have varied from 5.0% (in *Ameiva ameiva*) to 100% (in *Tropidurus cocorobensis*, *T. erythrocephalus,* and *T. hispidus*) (Cunha-Barros and Rocha, 2000; Rocha et al., 2008) (Table 3). The total infestation prevalence by *E. alfreddugesi* among the specimens analysed here was 32.14% (n = 18), which was significantly lower than rates reported in earlier studies of tropidurids infected by *E. alfreddugesi*, as for example *T. torquatus* (n = 146) from an area of *restinga* (sandy coastal) vegetation in Maricá, Rio de Janeiro State, with 97% prevalence (Cunha-Barros and Rocha, 2000); or *T. cocorobensis* (n = 16), *T. erythrocephalus* (n = 13), *T. hispidus* (n = 20), and *T. semitaeniatus* (n = 33) from an area of *caatinga* (dryland) vegetation in Morro do Chapéu, Bahia State, with prevalences ranging from 97-100% (Rocha et al., 2008); or *Tropidurus itambere* (n = 74) with a prevalence rate of 88.2%, *T. oreadicus* (n = 85) with 87.6% and *T. torquatus* (n = 16) with 65.2% in the Brazilian *cerrado* (savanna) (Carvalho et al., 2006). The present reported prevalence rate is only slightly higher than the lowest rate of 17.3% reported for *T. torquatus* (n = 13) (Carvalho et al., 2006).

The total mean infestation intensity by *E. alfreddugesi* (9.27 ± 1.83, 1-25) was comparatively low when compared to the mean infestation intensity of 164.9 ± 161.9 reported for *T. torquatus* (Cunha-Barros and Rocha, 2000), or 36.67 ± 41.09 reported for *T. itambere* (Carvalho et al., 2006).

The presence of *Geckobiella* sp. represents the first report of this parasite using *T. hispidus* as a host in northeastern Brazil. The infestation rate of this parasite in the present study (prevalence 50%, and mean intensity of infestation of 5.64 ± 1.28, 1-25) was found to be within the range previously published for the species *E. alfreddugesi* (Cunha-Barros and Rocha, 1995, 2000; Cunha-Barros et al., 2003; Carvalho et al., 2006; Rocha et al., 2008).

Our data showed that mean intensity rate infestation by *E. alfreddugesi* by *Geckobiella* sp. differed consistently. These differences may arise when some species are favoured in the occupation of particular mocrohabitats in the body of a host. In some cases the host body surface may be occupied by a considerable number of parasites but they differ greatly in infestation rates due to the particular specificity to some microhabitats (Bittencourt and Rocha, 2003).

Data indicated that adults of the sexes did not differ regarding the overall infection rates and the mean intensities of infestation. These results probably are due to the low selectivity of their parasites (Carvalho et al., 2006).

The sites most infested by *E. alfreddugesi* in the present study were the mite pockets, as was also found in *T. hispidus* in areas in Bahia state (Rocha et al., 2008). These pockets offer protection from mechanical shocks and from dehydration and may act to isolate the mites (Cunha-Barros and Rocha, 2000; Cunha-Barros et al., 2003; Garcia-de-la-Pena et al., 2004). Additionally, once the mites are established in these pockets they cannot be easily removed (Carvalho et al., 2006). However, in addition to the mite pockets, skin wrinkles (especially those found on the neck and inguinal regions) were the most highly

Table 3. Parasite species of different Brazilian lizard species (with their respective host habitat type), and values of prevalence (in %), intensity of infection, as well as the corresponding range of the infection intensity, and source of the data. Pa = Parasite specie; Pr = Prevalence; M = Mean Intensity.

Hosts	Habitats	N	Pa	Pr	M	References
Ameiva ameiva	Restinga	42	*E. alfreddugesi*	5.0%	1.0	Cunha-Barros and Rocha (2000)
Cnemidophorus littoralis	Restinga	100	*E. alfreddugesi*	72.0%	8.3 ± 10.2	Cunha-Barros and Rocha (2000)
	Restinga	21	*E. alfreddugesi*	95.2%	19.1 ± 16.8	Cunha-Barros et al. (2003)
Mabuya agilis	Restinga	26	*E. alfreddugesi*	96.1%	105.7	Cunha-Barros and Rocha (1995)
	Restinga	26	*E. alfreddugesi*	96.1%	110.1 ± 115.8	Cunha-Barros and Rocha (2000)
	Restinga	7	*E. alfreddugesi*	100%	20.9 ± 9.3	Cunha-Barros et al. (2003)
Mabuya macrorhyncha	Restinga	72	*E. alfreddugesi*	94.4%	42.6	Cunha-Barros and Rocha (1995)
	Restinga	78	*E. alfreddugesi*	94.0%	42.4 ± 50.3	Cunha-Barros and Rocha (2000)
	Restinga	12	*E. alfreddugesi*	100%	11.1 ± 13.1	Cunha-Barros et al. (2003)
Tropidurus cocorobensis	Ecotone(*)	16	*E. alfreddugesi*	100%	70.1 ± 41.7	Rocha et al. (2008)
Tropidurus erythrocephalus	Ecotone(*)	13	*E. alfreddugesi*	100%	165.8 ± 126.0	Rocha et al. (2008)
Tropidurus hispidus	Ecotone(*)	20	*E. alfreddugesi*	100%	146.2 ± 114.2	Rocha et al. (2008)
	Montane secondary forest	56	*E. alfreddugesi*	37.5%	10.2 ± 8.7	Present study
Tropidurus itambere	Cerrado	85	*E. alfreddugesi*	88.2%	36.67 ± 41.09	Carvalho et al. (2006)
Tropidurus oreadicus	Cerrado	97	*E. alfreddugesi*	87.6%	15.38 ± 21.08	Carvalho et al. (2006)
Tropidurus semitaeniatus	Ecotone(*)	34	*E. alfreddugesi*	97.1%	52.3 ± 42.4	Rocha et al. (2008)
Tropidurus torquatus	Restinga	146	*E. alfreddugesi*	97.7%	164.9 ± 161.9	Cunha-Barros and Rocha (2000)
	Restinga	62	*E. alfreddugesi*	100%	86.4 ± 94.6	Cunha-Barros et al. (2003)
	Cerrado	75	*E. alfreddugesi*	65.2%	12.13 ± 21.09	Carvalho et al. (2006)
Tropidurus hispidus	Montane secondary forest	56	*Geckobiella* sp.	50%	5.9 ± 6.8	Present study

infested sites in *T. itambere, T. oreadicus* and *T. torquatus* from the Cerrado (Carvalho et al., 2006); skin wrinkles were the preferred micro-habitats for mites among lizards captured in the restinga region at Maricá, perhaps because these animals have imbricate scales that also facilitate mite feeding and protection (Cunha-Barros and Rocha, 2000).

Scale patterns appear to influence rates of parasitism (Cunha-Barros and Rocha, 2000), as does the presence and the morphology of the mite pockets (Carvalho et al., 2006). Additionally, tropidurids show large variation in the number of pockets found among different species (Rodrigues, 1987). Considering the fact that *Tropidurus hispidus* has two pockets on its neck and two more in the axilar region (Rodrigues, 1987), and according to Clopton and Gold (1993) populations of *E. alfreddugesi* are sensitive to environmental variations and the effects of environmental degradation, the infestation rates of these lizards would be expected to differ from those observed in the present study.

Geckobiella sp. was found to be present on every part of the body in the lizards in the present study, with no specific preferred site, reflecting the fact that these mites are pterygossomatids that live under the imbricate scales of their hosts (Bertrand et al., 1995). Studies of infestations on Tropiduridae lizards undertaken in Brazil have previously identified only a single parasite, the trombicuilid *Eutrombicula alfreddugesi* (Cunha-Barros and Rocha, 2000; Cunha-Barros et al., 2003; Carvalho et al., 2006; Rocha et al., 2008). We presented here, however, evidence for two mite species infesting the same host. But, as the two mite species occupy distinct micro-habitats on the same host they can potentially avoid mutual competition - as has been seen with some species of nematodes found in specific infection sites in the digestive tracts of the same lizard species (see Bush et al., 2001). We did not find skin lesions on any of the infested specimens or any indication that these animals were debilitated or demonstrated behavioural modifications, but additional studies will be undertaken in the near future to more closely examine these aspects.

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