

FERNANDO DE CASTRO JACINAVICUS

**Chigger mites (Trombidiformes: Trombiculidae) of small mammals from Brazil:** morphological studies and investigation of the presence of pathogens

São Paulo

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**Chigger mites (Trombidiformes: Trombiculidae) of small mammals from Brazil: morphological studies and investigation of the presence of pathogens**

Thesis submitted to the Postgraduate Program in Experimental Epidemiology applied to Zoonoses of the School of Veterinary Medicine and Animal Science of the University of São Paulo to obtain the Doctor's degree in Sciences.

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*This thesis is dedicated to my wife  
Fernanda and my son Gabriel.*

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*“A time to be born, and a time to die; a time to plant,  
and a time to pluck up that which is planted”.*

Ecclesiastes 3:2

## RESUMO

JACINAVICIUS, FC. **Trombiculídeos (Trombidiformes: Trombiculidae) de pequenos mamíferos do Brasil:** estudos morfológicos e investigação da presença de patógenos. 2019. 148 p. Tese (Doutorado em Ciências) – Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo, 2019.

Trombiculídeo é o nome popular do estágio larval de ácaros da família Trombiculidae que parasitam vertebrados, e durante o parasitismo, podem causar uma reação cutânea no hospedeiro, conhecida como trombiculíase. Algumas espécies são vetores de tsutsugamushi, uma riquetsiose humana, causada pela bactéria *Orientia tsutsugamushi*. A presença de alguns patógenos tem sido detectada em ácaros trombiculídeos, como *Coxiella*, *Hepatozoon* e *Rickettsia*. Para o Brasil, apenas a bactéria *Rickettsia felis-like* foi detectada no ácaro da espécie *Blankaartia sinamaryi* coletada parasitando uma ave. Na região Neotropical, os trombiculídeos são pouco estudados e existem lacunas na taxonomia, biodiversidade e associação com hospedeiros. Considerando a carência de conhecimento, e que existem indícios da possibilidade de as larvas participarem da transmissão de patógenos em outras regiões do mundo, torna-se relevante a análise investigativa de *Coxiella*, *Hepatozoon* e *Rickettsia* neste grupo de ácaros no Brasil. Coleções nacionais e internacionais foram consultadas e os espécimes coletados parasitando mamíferos foram examinados. Larvas recentemente coletadas nos hospedeiros foram mantidas vivas, em colônias, para o conhecimento das ninfas e todos os estágios foram posteriormente utilizados em estudos moleculares e na detecção de patógenos. Ao final deste estudo, foram registradas 73 espécies de trombiculídeos, sendo que 17 espécies representam novos registros de localidade, dez foram relatadas pela primeira vez no país, deutoninha de uma espécie foi descrita, novas associações (24) de larvas parasitando mamíferos no Brasil foram fornecidas, duas novas espécies foram descritas e o gênero *Arisocerus* foi sinonimizado com o gênero *Herpetacarus*. Além disso, fornecemos o primeiro registro da detecção de *Rickettsia* sp. nos ácaros das espécies *A. hertigi*, *Q. trapezoides* e *T. bakeri* coletados em roedores no território nacional, incluindo a obtenção de sequências do gene 18S para as espécies *A. hertigi*, *Kymocta* sp., *Q. pazca*, *Q. trapezoides* e *T. bakeri* pela primeira vez, contribuindo com o banco de genes desse grupo de ácaros tão diverso mas ainda pouco conhecido no Brasil.

Palavras-chave: Trombiculídeos, taxonomia, 18S rDNA, Mammalia, Rickettsia

## ABSTRACT

JACINAVICIUS, FC. **Chigger mites (Trombidiformes: Trombiculidae) of small mammals from Brazil:** morphological studies and investigation of the presence of pathogens. 2019. 148 p. Tese (Doutorado em Ciências) – Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo, 2019.

Chiggers are the popular name of larval stage of mites of the family Trombiculidae that parasite vertebrates, and during the parasitism, can cause a skin reaction in the host, known as trombiculiasis. Some species are vectors of tsutsugamushi, a human rickettsiosis, caused by the bacteria *Orientia tsutsugamushi*. Furthermore, the presence of some pathogens has been detected in chiggers, as *Coxiella*, *Hepatozoon* and *Rickettsia*. To Brazil only the bacteria *Rickettsia felis*-like was detected in the chigger species *Blankaartia sinamaryi* collected parasitizing a bird. In the Neotropical region this group of mites is currently poorly studied and there are gaps in taxonomy, biodiversity, and association with vertebrate. Considering in the lack of knowledge, and that there are indications of the possibility of the larvae participating in the transmission of pathogens in other regions of the world, the investigative analysis of *Coxiella*, *Hepatozoon* and *Rickettsia* in this group of mites in Brazil becomes relevant. National and international collections were consulted and the specimens collected parasitizing mammals were examined. Besides that, chigger mites collected recently were kept in colonies for the knowledge of nymph stages and all stages were used for molecular studies and in pathogen detection. By the end of this study, for the national territory 73 species are recorded, 17 species have been recorded in new localities in Brazil, 10 species are reported for the first time in the country, the deutonymph stage for one species was described, 24 new associations of chiggers parasitizing mammals in Brazil have been provided, two new species were described and the genus *Arisocerus* was synonymized with the genus *Herpetacarus*. In addition, we provide the first report of the detection of strains of *Rickettsia* sp. in the chigger mites *A. hertigi*, *Q. trapezoides*, and *T. bakeri* collected on rodents in Brazil, including sequences of 18S gene of the species *A. hertigi*, *Kymocta* sp., *Q. pazca*, *Q. trapezoides*, and *T. bakeri* for the first time, increasing the genebank for this group of mites so diverse but still little known in Brazil

Keywords: Chiggers, taxonomy, 18S rDNA, Mammalia, *Rickettsia*.

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## 1. INTRODUCTION

The mites known as "chiggers", "chigger mites", "harvest mites" or "sand mites" belong to three families: Trombiculidae Ewing, 1944, Leeuwenhoekiidae Womersley, 1944, and Walchiidae Ewing, 1946 sensu Wen (1999). They can also be known as "thalzahuates" or "coloradillas" in Mexico, "red bugs" in England, "Herbstgrassmilben" in Germany and "tsutsugamushi" or "akamushi" in Japan (FONSECA, 1932; HOFFMANN, 1990). Aragão (1936) reported that the larvae of chiggers are easily confused with tick larvae and both receive the same generic name "Micuim". So, the author proposed to identify tick larvae as "micuim castanho" and chigger larvae as "micuim amarelo".

The larvae are ectoparasites of vertebrates, and during the parasitism, they liquefy and suck the host's epithelial tissue by the enzymatic action of their saliva. Blood is not ingested in this process, but some blood cells can be found in the gut of the mites (JONES, 1950). Deutonymphs and adults are free living predators (CROSSLEY, 1960).

During feeding of the larva, a tube known as stylostome (Figure 1) forms in the host skin, resulting from the salivary secretions of the mite and also from the immunological reactions of the host (HASE et al., 1978). The stylostome allows the parasite to reach the layers of deeper tissues of the skin (SHATROV, 2000), and individuals of some species may be wrapped by host tissue, remaining in capsules for up to six months (WHARTON; FULLER, 1952).



**Figure 1.** Microscopy image of the feeding tube (stylostoma) formed during the parasitism of *Quadrassetas brasiliensis* (IBSP 12706B) in *Euryoryzomys russtaus*.

Miyajima and Okumura (1917) were the first to provide information on the life cycle of chigger mites, studying the species *Leptotrombidium akamushi* (Brumpt, 1910). The authors consider seven stages: egg, prelarva or deutovum, larva, protonymph, deutonymph, tritonymph and adults, with larvae, deutoninfas and adults being active, while the other stages are quiescent (inactive). Ewing (1944) disregarded the inactive phases as properly "stages", named these phases as being, nothing more than, exticular cuticular shells.

Studies of the biology and taxonomy of these mites were further intensified, especially during World War II, increasing considerably the number of known species. American soldiers were attacked in trenches and encampments and contracted rickettsioses from infected chigger larvae in Burma and Sri Lanka (WHARTON; FULLER, 1952). According to these authors, public health departments were faced with the need to catalog, describe and know the biology of these mites. Takahashi et al. (2004) reported that larvae of some species of *Leptotrombidium* disseminate tsutsugamushi, a human rickettsiosis caused by *Orientia tsutsugamushi* and the rodents are natural reservoirs of this bacterium. The authors comment that the infected larva transmits the pathogen to the following stages (transestadial perpetuation) and to subsequent generations (transovarian transmission). Traub et al. (1975) and Kampen et al. (2004) stated that successful transmission of pathogens is only possible through one of these mechanisms.

In Europe, chigger mites of the genus *Neotrombicula* were naturally infected by bacteria *Anaplasma phagocytophilum* in Spain (FERNÁNDEZ-SOTO; PÉREZ-SÁNCHEZ; ENCINAS-GRANDES, 2001); *Borrelia burgdorferi* sensu lato in Germany and Czech Republic (KAMPEN et al., 2004; HUBÁLEK et al., 1998); and *B. garinii*. and *B. valaisiana* in Czech Republic (LITERAK et al., 2008). These authors suggest that chiggers are involved in some way in the epidemiology of borreliosis and anaplasmosis. In Africa, chigger mites were identified as potential cause of Q-fever by the transmission of *Coxiella burnetti* (DANIEL, 1961; KEPKA, 1965). Le Gac et al. (1953) reported a Q-fever case in a laboratory technician who had manipulated chigger larvae collected from an endemic area in Equatorial Africa. Blanc et al. (1952) successfully described *C. burnetti* infection in mites of the species *Neotrombicula autumnalis* (Shaw, 1790) in the laboratory. Mitkova et al. (2015) found two species of *Rickettsia*, *R. monacensis* and *R. helvetica* in the chigger species *Hirsutiella zachvatkini* (Schluger, 1948) and *R. monacensis* in *Kepkatrombicula storkani* (Daniel, 1956), in Slovakia. Recently, Weitzel et al. (2016) reported cases of exanthematic typhus caused by *O. tsutsugamushi* in people from the island of Chiloé, Chile, suggesting that chiggers could be vectors. Besides that, Frank (1977) detected the presence of the protozoa *Hepatozoon*

*erhardovae* in chigger of the species *Hirsutiella zachvatkini* (Schluger 1948), in Austria. Then, chigger mites may be considered potential vectors of pathogens; however the real role of these mites in the zoonosis epidemiology is still unknown in the Neotropical region (AZAD, BEARD, 1998; POINAR, POINAR, 1998).

In Brazil, as soon as the first cases of Brazilian Macular Fever (FMB) were diagnosed in the state of São Paulo, Fonseca (1932) emphasized the importance of hematophagous mites of the families Macropyssidae and Trombiculidae, as potential vectors. However, the role of these mites as vectors has not been confirmed. According to the author, there was an FMB outbreak, and 60 people were admitted to Emílio Ribas Hospital at that time, but only one had a tick fixed to the skin. In other cases, it was not possible to confirm the transmitting agent.

Ewing (1931) compiled the first list of chigger mites from the New World, eight species from Brazil. Wharton and Fuller (1952) increased the number by eight genera and 18 species. Fonseca (1955) reported trombiculiasis in human for Brazil. Brennan and Goff (1977) reported 87 chigger genera to the Nearctic and Neotropical regions, of which 18 genera were recorded from Brazil.

According to Pallini et al. (2007), the group of mites known as chiggers in Brazil was not nearly studied and little was known about taxonomy, biodiversity, association of vertebrate hosts and the possibility of pathogen detection. So, the approval of project CNPq 454907 / 2014-1 allowed the access and examination of types and material of chiggers deposited in national and international collections, as well as molecular studies of the presence of pathogens in recently collected mites. In addition, considering that there are indications of the possibility of chiggers participating in the transmission of pathogens in other regions of the world, the investigative analysis of *Rickettsia*, *Coxiella* and *Hepatozoon* in this group of mites in Brazil becomes relevant.

## 1.1 CHAPTER PRESENTATION

The results obtained in this study are organized in chapters. The chapter 1 is a checklist of chiggers that occur in Brazil, that were valid as of late 2017 (December 2017), including some new records for the country; the chapter 2 is about the knowledge of the larva and deutonymph stages of *Quadrasetas brasiliensis*; the chapter 3 comprises a description of a new species and new records of chiggers in Rio de Janeiro State, housed at the CAVAIIS-IOC collection; the chapter 4 is about chiggers from marsupials of the Pernambuco state, including a description of a new species and new records of localities. The

chapter 5 is a proposal of synonymization of genus *Arisocerus* with the genus *Herpetacarus*, including new records of species collected in Brazil; the chapter 6 is a result based on examination of Collections that housed samples of chiggers collected parasitizing mammals in Brazil; finally, the chapter 7 comprises the molecular results of chiggers submitted targeting pathogen detection. All the chapters are adapted in manuscript format, as follow:

- Chapter 1 -A checklist of chiggers from Brazil, including new records (Acari: Trombidiformes: Trombiculidae and Leeuwenhoekiidae). [Published on Zookeys 743: 1-41, 2018, doi: 10.3897/zookeys.743.22675].
- Chapter 2 - A contribution to the knowledge of *Quadrasetas brasiliensis* Goff and Gettinger, 1989 (Trombidiformes: Trombiculidae), with description of the deutonymph instar. [Published on Acarologia 58: 442-456, 2018, doi: 10.24349/acarologia/20184252].
- Chapter 3 - Description of *Parasacia fernandae* sp.n. (Trombidiformes: Trombiculidae) and new records of chiggers from rodents in Rio de Janeiro state, Brazil. [Published on Acarina 26: 205-211, 2018, doi: 10.21684/0132-8077-2018-26-2-205-211].
- Chapter 4 - Description of *Pseudoschoengastia petrolinensis* n. sp. (Trombidiformes: Trombiculidae), includes new records of chiggers from northeastern Brazil. [published on International Journal of Acarology, in press, 2019, doi: 10.1080/01647954.2019.1601766].
- Chapter 5 – Synonymy of the genus *Arisocerus* Brennan, 1970 with the genus *Herpetacarus* Vercammen-Grandjean, 1960 (Trombidiformes: Trombiculidae). [unpublished yet].
- Chapter 6 – New records of chiggers parasitizing mammals in Brazil housed at National and International Acari Collections. [unpublished yet].
- Chapter 7 - Molecular detection of pathogens in chigger mites (Trombidiformes: Trombiculidae) collected in small Brazilian mammals [unpublished yet].

## 1.2 GENERAL OBJECTIVE

To catalog the species of chiggers that parasitizes small mammals from Brazil, and to investigate the presence of pathogens associated.

### 1.2.1 Specific objectives

- Catalog the types and material of chiggers from Brazil deposited in the main national and international acarological collections;
- Detail and illustrate the morphology of the species examined, when possible.
- Investigate the presence of *Coxiella*, *Rickettsia* and *Hepatozoon*, in the mites recently collected.

### 1.3 REFERENCES

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## 2. CHAPTER 1 - A CHECKLIST OF CHIGGERS FROM BRAZIL, INCLUDING NEW RECORDS (ACARI: TROMBIDIFORMES: TROMBICULIDAE AND LEEUWENHOEKIIDAE)

### 2.1 INTRODUCTION

The mites known as “chiggers” or “chigger mites” belong to three families: Trombiculidae Ewing, 1944, Leeuwenhoekiidae Womersley, 1944, and Walchiidae Ewing, 1946 sensu Wen (1999). The larvae are ectoparasites of vertebrates, and during the parasitism, they liquefy and suck the host’s epithelial tissue by the enzymatic action of their saliva. Blood is not ingested in this process, but some blood cells can be found in the gut of the mite (Jones 1950). Deutonymphs and adults are free living predators (Crossley 1960).

Studies of the biology and taxonomy of these mites were intensified during World War II, thus increasing considerably the number of known species. Soldiers were attacked in encampments and contracted rickettsioses from infected larvae (Wharton and Fuller 1952). According to these authors, in the front of the situation, the public health departments were faced with the need for cataloging studies, description, morphology, and biology of these mites. Although they are considered potential vectors of pathogens, the role of these mites in the epidemiology of rickettsiosis is unknown in the Neotropical region (Azad and Beard 1998, Poinar and Poinar 1998).

In South America, the countries that have a checklist of chiggers species are: Venezuela (Brennan and Reed 1975), Peru (Brennan and Jones 1961), Surinam (Brennan and Lukoschus 1971) and recently Chile (Stekolnikov and González-Acuña 2015). Ewing (1931) compiled the first list of chigger mites from the New World, eight species from Brazil. Wharton and Fuller (1952) increased the number by eight genera and 18 species. Fonseca (1955) reported the first human hosts for Brazil. Brennan and Goff (1977) reported 87 chigger genera for the Nearctic and Neotropical regions, of which 18 were recorded from Brazil.

The Brazilian territory is composed by five regions (north, northeast, centralwest, southeast and south), 26 states and one federal district (Contel 2014). Between the 1970s and 1990s, new species were described by various researchers from specimens collected primarily in the northern and central-western parts of the country, but have never been summarized (Stekolnikov and González-Acuña 2015). Therefore, here we are compiling a checklist of species from Brazil, including new hosts and localities.

## 2.2 MATERIALS AND METHODS

In the checklist, the taxa comprise two families, *Trombiculidae* and *Leeuwenhoekiidae*. The genera and species are arranged alphabetically, in the follow format: Genus – Valid name, author and date; Species – Checklist number, valid name of species, author, date, type locality, host, and type depository, in parenthesis are the typification and the collection number, when available. In cases where the original host name has changed, the current host name is listed with the original host name in parenthesis. The host species were updated based on the IUCN (2017).

The following abbreviations were used: BPBM - USA, Hawaii, Honolulu, Bernice Pauahi Bishop Museum; FMNH - USA, Illinois, Chicago, Field Museum of Natural History Chicago (cited in original description as CNHM); IBSP Brazil, São Paulo, São Paulo, Instituto Butantan; MNRJ - Brazil, Rio de Janeiro, São Cristovão, Universidade do Rio Janeiro, Museu Nacional; MZSP - Brazil, São Paulo, São Paulo, Museu de Zoologia da Universidade de São Paulo; NHMUK - United Kingdom, London, The Natural History Museum [formerly British Museum (Natural History)] (cited in original description as BMNH or BM); OMNH - USA, Oklahoma, Norman, University of Oklahoma, Oklahoma Museum of Natural History; OSAL - USA, Ohio, Columbus, Ohio State University Acarology Laboratory; RMNH - Netherlands, Leiden, Naturalis Biodiversity Centre [formerly Rijksmuseum van Natuurlijke Historie]; SAM - Australia, South Australia, Adelaide, South Australian Museum; USNM - USA, Washington D.C., National Museum of Natural History, [formerly United States National Museum] which is housed at the Systematic Entomology Laboratory (BARC-USDA-ARS), cited in original descriptions as NMNH.

Details of the records in Brazil were provided based on the bibliographic review, followed to the new records for the country, when available. The new records from the checklist are based on the material deposited in the IBSP collection. Some of these species were also reported outside Brazilian territory. These recordings of literature were included in other records.

In addition, a list of chiggers from Brazil is summarized with their respective hosts (Appendix 1) and a list of vertebrate hosts and their respective chiggers and the published information from Brazil (Appendix 2).

## 2.3 RESULTS

A Brazilian checklist with eight species of the family Leeuwenhoekiidae and 55 of the family Trombiculidae, totaling 63 chigger species in 30 genera from approximately 70 different hosts is presented. Some regions just have one or two chigger records and most of these records are represented by the type series. Species of the family Walchiidae were never recorded in the Brazilian territory until this work.

The chigger species *P. aemulata*, *Q. flochi*, *Q. mirandae*, and *Q. trapezoides* are reported for the first time in Brazil, and new records of locality for *T. bakeri* and new localities and hosts for the species *C. spinosus*, *E. tinami*, *K. brasiliensis*, *Q. pazca*, and *S. tamarina* are provided too.

### 2.3.1 Checklist of larvae chigger species (in alphabetical order) recorded from Brazil

#### Family LEEUWENHOEKIIDAE Womersley, 1944

##### Genus *Apolonia* Torres and Braga, 1938: 172

1. *Apolonia tigipioensis* Torres and Braga, 1938: 172; Tigipió, Pernambuco state, Brazil, ex *Gallus gallus domesticus*; IBSP (syntype, n° 1954).

**Records from Brazil.** In addition to the type data, Carneiro (1949, 1952) reported *A. tigipioensis* parasitizing humans and *Nothura maculosa cearensis* (Tinamiformes) in Limoeiro (Pernambuco). This chigger was also found parasitizing the birds *Struthio camelus* (Struthioniformes) and *Passer domesticus* (Passeriformes) in Petrolina (Pernambuco) (Ornelas-Almeida et al. 2007).

**Other records.** Venezuela (Brennan and Reed 1975).

##### Genus *Hannemania* Oudemans, 1911: 137

2. *Hannemania hepatica* Fonseca, 1936: 30; Butantan, São Paulo, São Paulo state, Brazil; ex. *Leptodactylus latrans* (= *L. ocellatus*); IBSP (holotype, n° 31).

**Records from Brazil.** Only the type data.

3. *Hannemania hylodeus* (Oudemans, 1910): 88; Brazil, ex *Hylodes* sp.; RMNH (nº 6247).

**Records from Brazil.** Only the type data, no locality information was provided by the author.

4. *Hannemania newsteadi* Sambon, 1928: 127; Urucum, Mato Grosso do Sul state, Brazil, ex *Hyla rubra*; NHMUK.

**Records from Brazil.** Only the type data.

5. *Hannemania stephensi* Sambon, 1928: 127; Tombador, Mato Grosso state, Brazil, ex *Pristimantis conspicillatus* (= *Hylodes conspicillatus*); NHMUK.

**Records from Brazil.** Only the type data.

#### Genus *Leeuwenhoekia* Oudemans, 1911: 137

6. *Leeuwenhoekia verduni* (Oudemans, 1910): 88; southern Brazil; ex. *Didelphis* sp., “opossum” RMNH (nº 6250).

**Records from Brazil.** Only the type data, this species was described from an unknown location in southern Brazil.

#### Genus *Whartonias* Ewing, 1944: 102

7. *Whartonias nudosetosa* (Wharton, 1938): 142; Cave near Oxkutzcab, Yucatan, Mexico, ex *Artibeus jamaicensis yucatanicus* and “*Pteropteryx canina canina*”; USNM (nº 1264).

**Note:** The correct host name is *Pteropteryx macrotis*, originally described as *Verpertilio caninus*.

**Records from Brazil.** Silveira et al. (2015) reported this species from Medina and São José da Safira (Minas Gerais state) on *Carollia perspicillata* (Chiroptera).

**Other records.** Costa Rica (Webb and Loomis 1977); Guatemala (Brennan and Dalmat 1960); Jamaica (Brennan 1953); Nicaragua (Webb and Loomis 1977); Mexico (Hoffmann 1949; Loomis 1969; Webb and Loomis 1977; Wharton 1938); Surinam (Brennan and van Bronswijk 1975); Trinidad (Brennan and Jones 1960, Brennan 1967) and Venezuela (Reed and Brennan 1975).

8. *Whartonias pachywhartoni* Vercammen-Grandjean, 1966: 282; Lagoa Santa, Minas Gerais state, Brazil, ex *Micronycteris megalotis*, USNM (holotype, nº 10462/6).

**Records from Brazil.** In addition to the type data, *W. pachywhartoni* was found in São José da Safira (Minas Gerais) parasitizing the bat *C. perspicillata* (Silveira et al. 2015).

#### Family TROMBICULIDAE Ewing, 1944

##### Genus *Aitkenius* Brennan, 1970; 1694

9. *Aitkenius vellosus* Brennan, 1970: 1695; Belém, Pará state, Brazil, ex *Proechimys guyannensis*, USNM (holotype and paratypes, nº 48876), FMNH (paratypes) and NHMUK (paratypes).

**Records from Brazil.** In addition to the type data, *A. vellosus* was also found at the same locality, on *Hylaeamys megacephalus* (Rodentia), and in Bragança (Pará) on the water rat *Nectomys squamipes* (Rodentia) (Brennan 1970d).

##### Genus *Arisocerus* Brennan, 1970: 32

10. *Arisocerus amapensis* Brennan, 1970; 32; Serra do Navio, Amapá state, Brazil, ex *Euryoryzomys macconnelli*; USNM (holotype and paratypes, nº 49368).

**Records from Brazil.** In addition to the type data, this species was also found at the same locality, on *H. megacephalus*, and in Belém (Pará) parasitizing *H. megacephalus* and *P. guyannensis* (Brennan 1970c).

**Other records.** Suriname (Brennan 1970c, Brennan and van Bronswijk 1975) and Venezuela (Brennan and Reed 1975).

11. *Arisocerus hertigi* (Brennan and Jones, 1964): 308; Sommerfield, Paraguay, ex “agouti”; USNM (holotype and paratypes, nº 43427), FMNH (paratypes), OSAL (paratypes), BPBM (paratypes).

**Records from Brazil.** This species was found in Brasília (Federal District), parasitizing the marsupial *Didelphis albiventris* (Didelphimorphia) (Goff and Gettinger 1989). In addition, *A. hertigi* was found on the Archipelago Moleques do Sul (Santa Catarina) parasitizing *Cavia intermedia* (Rodentia) (Regolin et al. 2015).

**Other records.** Paraguay (Brennan and Jones 1964a).

**Genus *Blankaartia* Oudemans, 1911: 123**

**12.** *Blankaartia shatrovi* Bassini-Silva and Barros-Battesti, 2016: 83; São Paulo, São Paulo state, Brazil, ex “bird”; IBSP (holotype and paratypes, nº 367).

**Records from Brazil.** In addition to the type data, this species was also found in Coronel Pacheco (Minas Gerais) parasitizing *Trichothraupis melanops* (Passeriformes) (Bassini-Silva et al. 2016).

**13.** *Blankaartia sinnamaryi* Floch and Fauran, 1956: 3; Sinnamary, French Guiana, ex *Homo sapiens*.

**Records from Brazil.** This species was found in Paraty (Rio de Janeiro) parasitizing *Manacus manacus* (Passeriformes), in Juiz de Fora (Minas Gerais) parasitizing *Picumnus cirratus* (Piciformes), *Tachyphonus coronatus*, *Thamnophilus caerulescens* and *Turdus rufiventris* (Passeriformes), and in Coronel Pacheco (Minas Gerais) parasitizing *Conopophaga lineata* and *Turdus albicollis* (Passeriformes) (Bassini-Silva et al. 2016).

**Other records.** Costa Rica (Arnold 1970, Stekolnikov et al. 2007), Cuba (Daniel and Stekolnikov 2003), French Guiana (Floch and Fauran 1956), Jamaica (Brennan 1953), Panama (Brennan and Yunker 1966), Peru (Brennan and Jones 1961a), Surinam (Brennan and van Bronswijk 1975), Trinidad (Brennan and Jones 1960), USA (Brennan 1965, Spalding et al. 1997) and Venezuela (Brennan and Reed 1975).

**Genus *Buclypeus* Brennan, 1972: 1178**

**14.** *Buclypeus catatonus* Brennan, 1972: 1178; Belém, Pará state, Brazil, ex *Proechimys guyannensis*; USNM (holotype and paratypes, nº 49322), NHMUK (paratypes), BPBM (paratypes) and FMNH (paratypes).

**Records from Brazil.** Only the type data.

**15.** *Buclypeus ignotus* (Brennan, 1971): 214; Belém, Pará state, Brazil, ex *Proechimys guyannensis*, USNM (holotype and paratypes, nº 49322), NHMUK (paratypes) and FMNH (paratypes).

**Records from Brazil.** Only the type data.

**Genus *Caamembecaia* Gazêta, Amorim, Bossi, Linhares and Serra-Freire, 2006: 137**

**16.** *Caamembecaia gratusus* **Gazêta, Amorim, Bossi, Linhares and Serra-Freire, 2006:** 137; Itatiaia National Park, Rio de Janeiro state, Brazil, ex *Trinomys gratusus*; MNRJ (holotype, n° 67498).

**Records from Brazil.** Only the type data.

**Genus *Chiroptella* Vercammen-Grandjean, 1960: 469**

**17.** *Chiroptella (Oudemansidium) australis* **(Brennan, 1970):** 810; Pedras Negras, Rondônia state, Brazil, ex *Nyctinomops laticaudatus* (= *Tadarida laticaudata*); USNM (holotype and paratypes, n° 50192), NHMUK (paratypes) and FMNH (paratypes).

**Records from Brazil.** Only the type data.

**Other records.** Venezuela: (Brennan and Reed 1975).

**Genus *Colicus* Brennan, 1970: 271**

**18.** *Colicus brasiliensis* **Goff, Whitaker and Dietz, 1983:** 185; São Roque de Minas, Minas Gerais state, Brazil, ex *Oligoryzomys formesi* (= *Oryzomys formesi*) and *Cerradomys subflavus* (= *Oryzomys subflavus*); BPBM (holotype, n° 12711 and paratypes).

**Records from Brazil.** Only the type data.

**19.** *Colicus icomi* **Brennan, 1970:** 271; Serra do Navio, Amapá state Brazil, ex *Proechimys guyannensis*; USNM (holotype and paratypes, n° 49342).

**Records from Brazil.** Only the type data.

**Other records.** Surinam (Brennan and van Bronswijk 1975).

**20.** *Colicus spinosus* **Goff and Gettinger, 1989:** 554; Brasília, Federal District, Brazil, ex *Gracilinannus agilis*; MZSP (holotype), BPBM (paratypes), OMNH (paratypes) and USNM (paratypes).

**Records from Brazil.** Only the type data.

**New record.** IBSP 11116 (1 larva), Condomínio Vila Verde, Itapevi, São Paulo, state of São Paulo, 29-XI-2012, *Monodelphis* sp. (Didelphimorphia) (#VV 105).

**Genus *Euschoengastia* Ewing 1938: 293**

**21.** *Euschoengastia trouessarti* (Oudemans, 1910): 87; southern Brazil; ex *Didelphis* sp.; RMNH.

**Records from Brazil.** Only the type data, this species was described from an unknown location in southern Brazil.

### Genus *Eutrombicula* Ewing, 1938: 293

**22.** *Eutrombicula alfreddugesi* Oudemans, 1910: 84; México, ex *Homo sapiens*; RMNH.

**Records from Brazil.** This species was found in Correntes (Mato Grosso) parasitizing the snake, *Xenodon merremii* and a human in São Paulo (São Paulo) (Fonseca 1932a). This species was also found in Unaí (Minas Gerais) and Pirenópolis (Goiás) parasitizing *Tropidurus oreadicus*, *Tropidurus itambere* and *Tropidurus torquatus* (Squamata) (Carvalho et al. 2006). There are additional records from Chapada do Araripe (Ceará) on *Tropidurus hispidus* (Squamata) (Delfino et al. 2011) and from Morro do Chapéu (Bahia), on *Tropidurus hispidus*, *Tropidurus cocorobensis*, *Tropidurus semitaeniatus* and *Tropidurus erythrocephalus* (Menezes et al. 2011). Recently this species was found parasitizing goats in Maranhão state (Faccini et al. 2017).

**Other records.** Bolivia (Brennan 1970a); Costa Rica (Arnold 1970); Cuba (Daniel and Stekolnikov 2004); USA (Rohani and Cromroy 1979); Guatemala (Brennan and Dalmat 1960); Mexico (Estébanes-González and Cervantes 2005, Hoffmann 1949, 1990, Jenkins 1949, Kroman et al. 1967, Loomis 1969, Loomis and Spath 1969, Oudemans 1910, Pearse et al. 1936, Wharton 1938); Panama (Brennan and Yunker 1966); Surinam (Brennan and van Bronswijk 1975; Brennan and Lukoschus 1971); Trinidad (Brennan and Jones 1960) and Venezuela (Brennan and Reed 1974).

**23.** *Eutrombicula batatas* (Linnaeus, 1758): 617; Surinam, ex unknown host; NHMUK.

**Records from Brazil.** This species was found in Manaus and Carvoeiro (Amazonas) parasitizing *G. domesticus*, and in Belém (Pará), parasitizing *Meleagris gallopavo* (Galliformes) (Ewing 1925). There are also records of *E. batatas* parasitizing *Dasyprocta agouti* (Rodentia) and unidentified Cervidae in Pará state (Bequaert 1926). Confalonieri and De Carvalho (1973) found *E. batatas* parasitizing *G. domesticus* in Mangaratiba (Rio de Janeiro). This species was also found parasitizing equines in Cáceres (Mato Grosso) (Confalonieri and Benez 1976) and on goats in Maranhão state (Faccini et al. 2017).

**Other records.** Argentina (Brennan and Jones 1964b); Bolivia (Brennan 1970a); Costa Rica (Arnold 1970); Curaçao (Brennan 1967); Guatemala (Brennan and Dalmat 1960); Jamaica (Brennan 1953); Mexico (Estébanes-González and Cervantes 2005, Hoffmann 1990, Jenkins 1949, Linnaeus 1758, Loomis 1969, Loomis and Stephens 1962); Panama (Brennan and Yunker 1966); Surinam (Brennan and van Bronswijk 1975, Brennan and Lukoschus 1971, Linnaeus 1758); Trinidad (Bennett and Loomis 1980, Brennan and Jones 1960) and Venezuela (Brennan and Reed 1974).

**24. *Eutrombicula bruyanti* (Oudemans, 1910): 85;** southern Brazil; ex *Didelphis* sp. “opossum”; RMNH.

**Records from Brazil.** Only the type data, this species was described from an unknown location in southern Brazil.

**25. *Eutrombicula goeldii* (Oudemans, 1910): 84;** Brazil; ex *Dasyprocta aguti*; RMNH.

**Records from Brazil.** Only the type data, no locality information was provided by the author.

**Other records.** Bolivia (Brennan 1970a); Colombia (Boshell and Kerr 1942, Brennan 1968a); Costa Rica (Arnold 1970); Dominica (Brennan 1967); Panama (Brennan and Yunker 1966); Surinam (Brennan 1970c, Brennan and van Bronswijk 1975, Brennan and Lukoschus 1971); Trinidad (Brennan and Jones 1960); Venezuela (Brennan and Reed 1974).

**26. *Eutrombicula tinami* (Oudemans, 1910): 84;** Brazil; ex *Crypturus noctivagus*; RMNH.

**Records from Brazil.** Only the type data, no locality information was provided by the author.

**New record.** IBSP 11368 (8 larvae), Condomínio Vila Verde, Itapevi, São Paulo, state of São Paulo, 19-VI-2013, *Didelphis aurita* (#VV 107).

**Other records.** Panama (Brennan and Lukoschus 1971); Surinam (Brennan and Lukoschus 1971); and Venezuela (Brennan and Reed 1974).

#### Genus *Fonsecia* Radford, 1942: 56

**27. *Fonsecia ewingi* (Fonseca, 1932): 153;** Correntes, Mato Grosso state, Brazil, ex *Xenodon merremii*; IBSP (syntypes, nº 27).

**Records from Brazil.** In addition to the type data, this species was also found at the following localities: Quiteriozinho (Mato Grosso), Birigui, Guatapará, Jacaré, Matão,

Penápolis, Promissão and Silvania (São Paulo), Morrinhos (Goiás) and Rio Negro (Paraná) (Fonseca 1932b).

**Other records.** Trinidad (Brennan 1967).

**28.** *Fonsecia ophidica* (Fonseca, 1932): 151; Promissão, São Paulo state, Brazil, ex *Xenodon merremii*; IBSP (syntypes, nº 29).

**Records from Brazil.** In addition to the type data, this species was also found in Matão (São Paulo), on the same host (Fonseca 1932b).

**29.** *Fonsecia travassosi* (Fonseca, 1936): 29; Angra dos Reis, Rio de Janeiro state, Brazil; ex *Spilotes pullatus*; IBSP (holotype, nº 30).

**Records from Brazil.** Only the type data.

#### Genus *Hooperella* Vercammen-Grandjean, 1967: 853

**30.** *Hooperella spinirostra* (Vercammen-Grandjean, 1967): 854; Lagoa Santa, Minas Gerais state, Brazil, ex *Micronycteris megalotis*; USNM (holotype, nº 10462/B).

**Records from Brazil.** Only the type data.

#### Genus *Kymocta* Yunker and Brennan, 1962: 572

**31.** *Kymocta brasiliensis* (Fonseca, 1936): 32; Butantan, São Paulo, São Paulo state, Brazil, ex “wild mouse”; IBSP (holotype, nº 334).

**Records from Brazil.** Only the type data.

**New records.** IBSP 11129C (3 larvae), Morro Grande, Cotia, state of São Paulo, 26-IV-2012, *Akodon montensis* (#MGR 38).

**32.** *Kymocta faitkeni* Brennan, 1968: 614; Serra do Navio, Amapá state, Brazil, ex *Hylaeamys megacephalus* (= *Oryzomys capito*); USNM (holotype and paratypes, nº 48875) and NHMUK (paratypes).

**Records from Brazil.** In addition to the type data, this species was record in the same locality and host by Brennan and van Bronswijk (1973) and was also found in Bragança (Pará) parasitizing *Necromys lasiurus* (= *Zygodontomys lasiurus*) (Rodentia) and *Didelphis marsupialis* (Didelphimorphia) (Brennan and van Bronswijk 1973).

**Other records.** Trinidad (Brennan 1968b) and Venezuela (Brennan and van Bronswijk 1973; Brennan and Yunker 1969).

**33. *Kymocta inca* (Brennan and Jones, 1961): 177;** Quince Mil, Cuzco, Peru, *Nephelomys keaysi* (= *Oryzomys keaysi*); USNM (holotype and paratypes, nº 33554).

**Records from Brazil.** In addition to the type data, this species was record in the same locality and host by Brennan and van Bronswijk (1973) and on the rodent *P. guyannensis* in Bragança (Pará) (Brennan and van Bronswijk 1973).

**Other records.** Peru (Brennan and Jones 1961a) and Venezuela (Brennan and Yunker 1969).

**34. *Kymocta lutui* Goff, Whitaker and Dietz, 1983: 185;** National Park of Serra da Canastra, Minas Gerais state, Brazil, *Necromys lasiurus* (= *Zygodontomys lasiurus*); BPBM (holotype and paratypes, nº 12712).

**Records from Brazil.** Only the type data.

#### Genus *Microtrombicula* Ewing, 1950: 297

**35. *Microtrombicula brachytrichia* Brennan, 1971: 214;** Belém, Para state, Brazil, *Proechimys guyannensis*; USNM (holotype and paratypes nº 50350).

**Records from Brazil.** Only the type data.

**36. *Microtrombicula brennani* Goff, Whitaker and Dietz, 1986: 171;** Poço das Antas Biological Reserve, Rio de Janeiro state, Brazil, ex *Lentopithecus rosalia*; USNM (holotype and paratypes) and BPBM (paratypes).

**Records from Brazil.** Only the type data.

**37. *Microtrombicula rhipidomysi* Goff, Whitaker and Dietz, 1983: 183;** National Park of Serra da Canastra, state of Minas Gerais, Brazil, ex *Rhipidomys mastacalis*; BPBM (holotype, nº 12710 and paratypes)

**Records from Brazil.** Only the type data.

**Genus *Neoschoengastia* Ewing, 1929: 187**

38. *Neoschoengastia esorrhina* Brennan, 1971: 666; Ananindeua, Pará state, Brazil, ex *Automolus infuscatus*, USNM (holotype and paratypes, n° 56193).

**Records from Brazil.** Only the type data.

**Genus *Parascoshoengastia* Vercammen-Grandjean, 1960: 469**

39. *Parascoschoengastia aemulata* (Brennan and Jones, 1964): 307; Rancho Grande, Aragua, Venezuela, ex *Anoura caudata*; USNM (holotype and paratypes, n° 40658), FMNH (paratypes), OSAL (paratypes), BPNM (paratypes).

**Records from Brazil.** Present study.

**New record.** IBSP 12558 (1 larva), Morro Grande, Cotia, São Paulo, 20-X-2015, *Akodon* sp. (#MGR 246).

**Other records.** Venezuela (Brennan and Jones 1964a).

**Genus *Parasecia* Loomis, 1966: 191**

40. *Parasecia aitkeni* (Brennan and Jones, 1960): 510; Cumaca, Trinidad, ex *Nectomys squamipes*; USNM (holotype and paratypes, n° 33628), FHMUK (paratypes), FMNH (paratypes) and SAM (paratypes).

**Records from Brazil.** In addition to the type data, this species was found in the same locality, but parasitizing the marsupial *Monodelphis domestica* (Whitaker and Dietz 1987).

**Other records.** Bolivia (Brennan 1970a); Surinam (Brennan and Lukoschus 1971); Trinidad (Brennan 1969c, Brennan and Jones 1960) and Venezuela (Brennan and Reed 1975).

41. *Parasecia fundata* Brennan, 1969: 664; Belém, Pará state, and Serra do Navio, Amapá state, Brazil, ex *Caluromys philander*, *Didelphis marsupialis* and *Glyphorynchus spirurus*; USNM (holotype and paratypes, n° 49323) and NHMUK (paratypes).

**Records from Brazil.** In addition to the type data, this species was also found parasitizing *P. guyannensis* in Serra do Navio (Amapá) (Brennan 1969c).

**Other records.** Costa Rica (Stekolnikov et al. 2007).

**42.** *Parasacia lasiurus* Goff and Gettinger, 1991: 401; Brasília, Federal District, Brazil, ex *Necromys lasiurus* and *Oxymycterus* sp.; MZSP (holotype), BPBM (paratypes), OMNH (paratypes).

**Records from Brazil.** Only the type data.

**43.** *Parasacia orphana* Brennan, 1971: 212; Belém, Pará state, Brazil, ex *Proechimys guyannensis*; USNM (holotype and paratypes, nº 50351), NHMUK (paratypes), FMNH (paratypes).

**Records from Brazil.** Only the type data.

**44.** *Parasacia thalurania* Brennan, 1969: 663; Belém, Pará state, Brazil; ex *Thalurania furcata*; USNM (holotype, nº 49374).

**Records from Brazil.** Only the type data.

**45.** *Parasacia valida* Brennan, 1969: 663; Bragança, Pará state, and Serra do Navio, Amapá state, Brazil, ex *Hylaeamys megacephalus*, *Necromys lasiurus* (= *Zygodontomys lasiurus fuscinus*) and *Monodelphis brevicaudata*; USNM (holotype and paratypes, nº 49092) and NHMUK (paratypes).

**Records from Brazil.** Only the type data.

**Other records.** Surinam (Brennan and Lukoschus 1971).

#### Genus *Paratrombicula* Goff and Whitaker, 1984: 329

**46.** *Paratrombicula plaumanni* (Brennan and Jones, 1964): 309; Nova Teutônia, Santa Catarina state, Brazil, ex *Ctenomys minutus*; USNM (holotype and paratypes, nº 34911), NHMUK (paratypes), FMNH (paratypes), OSAL (paratypes), BPNM (paratypes).

**Records from Brazil.** Only the type data.

**Other records.** Chile (Stekolnikov and González-Acuña 2012).

#### Genus *Perissopalla* Brennan and White, 1960: 303

**47.** *Perissopalla barticonycteris* Brennan, 1969: 429; Belém, Pará state, Brazil, ex *Glyphonycteris daviesi*; USNM (holotype and paratypes, nº 49939).

**Records from Brazil.** Only the type data.

**Other records.** Bolivia (Brennan 1970a) and Surinam (Brennan and van Bronswijk 1975, Brennan and Lukoschus 1971).

**48. *Perissopalla ipeani* Brennan, 1969: 429;** Belém, Pará state, Brazil, ex *Carollia perspicillata*; USNM (holotype and paratypes, nº 49366).

**Records from Brazil.** In addition to the type data, this species was also reported from *Tonatia bidens* (Chiroptera) in Parque Estadual Pedra Branca (Rio de Janeiro) (Almeida et al. 2011).

**Other records.** Surinam (Brennan and van Bronswijk 1975, Brennan and Lukoschus 1971).

**49. *Perissopalla tanydera* Brennan, 1969: 430;** Icabarú, Bolívar, Venezuela, ex *Peropteryx* sp.; RML (holotype and paratypes, nº 49833).

**Records from Brazil.** This species was found in Príncipe de Beira (Rondônia) parasitizing the bat *Saccopteryx bilineata* (Brennan 1970a).

**Other records.** Bolivia (Brennan 1970a); Surinam (Brennan and van Bronswijk 1975) and Venezuela (Brennan 1969b, Brennan and Reed 1975).

#### **Genus *Polylopodium* Brennan and Jones, 1961: 112**

**50. *Polylopodium asprium* Brennan, 1969: 868;** Serra do Navio, Amapá state and Bragança, Pará state, Brazil; ex *H. megacephalus* (= *Oryzomys capito*); RML (holotype and paratypes, nº 49341).

**Records from Brazil.** In addition to the type data, this species was found in the same host, but in Belém (Pará) (Brennan 1969a), and also was found on the rodent *Proechimys* sp. in Príncipe da Beira (Rondônia) (Brennan 1970a).

**Other records.** Venezuela (Brennan and Reed 1975).

#### **Genus *Quadrasetas* Brennan, 1970: 1695**

**51. *Quadrasetas brasiliensis* Goff and Gettinger, 1989: 557;** Brasília, Federal District, Brazil, ex *Hylaeamys megacephalus*, *Gracilinannus agilis*, *Monodelphis Americana*; MZSP (holotype), BPBM (paratypes) and OMNH (paratypes).

**Records from Brazil.** Only the type data.

**52.** *Quadraseta flochi* (Brennan and Jones, 1960): 503; Maingot state, Trinidad, ex *Rattus rattus*; USNM (holotype and paratypes, nº 33885), FMNH (paratypes), NHMUK (paratypes) and SAM (paratypes).

**Records from Brazil.** Present study.

**New record.** IBSP 11096F (2 larvae), Morro Grande, Cotia, São Paulo state, 20-VI-2012, *Euryoryzomys russatus* (#MGR 32).

**Other records.** Colombia (Brennan 1968a), Surinam (Brennan and Lukoschus 1971), Trinidad (Brennan and Jones 1960) and Venezuela (Brennan and Reed 1975).

**53.** *Quadraseta mirandae* Goff and Brennan, 1977: 504; Quebrada Chacaito, Miranda, Venezuela, ex *Nephelomys albicularis* (= *Oryzomys albicularis*); USNM (holotype and paratypes, nº 52518) and BPBM (paratypes).

**Records from Brazil.** Present study.

**New records.** IBSP 10606C (1 larva), Morro Grande, Cotia, São Paulo state, 13-IX-2011, *Akodon montensis* (#MGR 6); 1 larva (IBSP 11129B), same locality and host (#MGR 38), 26-VI-2012.

**Other records.** Venezuela (Goff and Brennan 1977).

**54.** *Quadraseta pazca* (Brennan and Jones, 1964): 700; Azul, Buenos Aires, Argentina, ex *Rattus* sp. and *Mus* sp.; USNM (holotype and paratypes, nº 38090), NHMUK (paratypes), FMNH (paratypes) and OSAL (paratypes).

**Records from Brazil.** Whitaker and Dietz (1987) reported this species from the Serra da Canastra National Park (Minas Gerais) on the rodent *Calomys tener*.

**New records.** IBSP 10606B (1 larva), Morro Grande, Cotia, São Paulo state, 13-IX-2011, *Akodon montensis* (#MGR 6); 2 larvae (IBSP 11096E), same locality, 20-VI-2012, *Euryoryzomys russatus* (#MGR 32).

**Other records.** Argentina (Brennan and Jones 1964b).

**55.** *Quadraseta trapezoides* (Brennan and Jones, 1964): 699; Azul, Buenos Aires, Argentina, ex “fox”; USNM (holotype and paratypes, nº 38095), NHMUK (paratypes), FMNH (paratypes) and OSAL (paratypes).

**Records from Brazil.** Present study.

**New record.** IBSP 11110C (12 larvae), Morro Grande, Cotia, São Paulo state, 16-VII-2012, *Nectomys squamipes* (#MGR 40).

**Other records.** Argentina (Brennan and Jones 1964b).

**Genus *Rhinibius* Brennan and Yunker, 1969: 304**

**56.** *Rhinibus tamandua* **Brennan and Yunker, 1969: 304;** Belém, Pará state, ex *Tamandua tetradactyla*; USNM (holotype and paratypes, nº 49365), FMNH (paratypes) and NHMUK (paratypes).

**Records from Brazil.** Only the type data.

**Other records.** Venezuela (Brennan and Reed 1975, Brennan and Yunker 1969).

**Genus *Serratacarus* Goff and Whitaker, 1984: 162**

**57.** *Serratacarus dietzi* **Goff and Whitaker, 1984: 163;** Serra da Canastra National Park, Minas Gerais state, Brazil; ex *Necromys lasiurus*; USNM (holotype and paratypes) and BPBM (paratypes).

**Records from Brazil.** Only the type data.

**58.** *Serratacarus lasiurus* **Goff and Whitaker, 1984: 166;** Serra da Canastra National Park, Minas Gerais state, Brazil; ex *Necromys lasiurus*; USNM (holotype and paratypes) and BPBM (paratypes).

**Records from Brazil.** Only the type data.

**Genus *Speleocola* Lipovsky, 1952: 132**

**59.** *Speleocola tamarina* **Goff, Whitaker and Dietz, 1987: 198;** Poço das Antas Biological Reserve, Rio de Janeiro state, Brazil, ex *Lentopithecus rosalia*; USNM (holotype and paratypes).

**Records from Brazil.** The type data and the present study.

**New records.** IBSP 11364B (1 larva), Morro Grande, Cotia, São Paulo, 21-VII-2013, *Sooretamys angouya* (#MGR 148).

### **Genus *Trombewingia* Fonseca, 1955: 3**

**60.** *Trombewingia bakeri* (Fonseca, 1955): 3; Reserva Florestal do Horto Florestal de São Paulo, São Paulo state, Brazil, ex *Guerlinguetus ingrami*; IBSP (Lectotype and Paralectotypes, nº 344).

**Records from Brazil.** In addition to the type data, Whitaker and Dietz (1987) reported it from the Serra da Canastra National Park (Minas Gerais) parasitizing the marsupial *M. domestica*. Other recent reports from São Paulo state include the municipality of Campos do Jordão and municipality of Cotia parasitizing *A. montensis*, and the rodents *Delomys dorsalis* and *Sooretamys angouya* (Jacinavicius et al. 2015).

**61.** *Trombewingia brasiliensis* Goff and Gettinger, 1991: 401; Brasília, Federal District, Brazil, ex *Necromys lasiurus* (= *Bolomys lasiurus*); MZSP (holotype), BPBM (paratypes) and OMNH (paratypes).

**Records from Brazil.** Only the type data.

### **Genus *Trombicula* Berlese, 1905: 155**

**62.** *Trombicula truncata* Brennan, 1970: 272; Príncipe de Beira, Rondônia state, Brazil, ex *Saccopteryx bilineata*; USNM (holotype and paratypes, nº 50188), FMNH (paratypes) and NHMUK (paratypes).

**Records from Brazil.** Only the type data.

### **Genus *Vercammenia* Audy and Nadchatram, 1957: 95**

**63.** *Vercammenia yorkei* (Sambon, 1928): 119; Urucum, Mato Grosso do Sul state, Brazil, ex *Scinax funereus* (= *Hyla rubra*); NHMUK (holotype, nº 147-9).

**Records from Brazil.** Only the type data.

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**3. CHAPTER 2 - A CONTRIBUITION TO THE KNOWLEDGE OF *Quadrasetas brasiliensis* GOFF AND GETTINGER, 1989 (TROMBIDIFORMES: TROMBICULIDAE), WITH DESCRIPTION OF THE DEUTONYMPH INSTAR**

**3.1 INTRODUCTION**

Around 3,000 species of chigger mites (Trombidiformes: Trombiculidae) have been described worldwide (Goff et al. 1982), and most of this diversity is known only by the type specimens. The life cycle of chiggers includes seven stages, namely egg, pre-larva, larva, protonymph, deutonymph, tritonymph and adult, yet active stages are larvae, deutonymphs and adults. While the majority of larvae from this group of mites are ectoparasites that feed on terrestrial vertebrates, other active stages are arthropod predators. Because of their parasitism in larval stage, the taxonomy of these mites has been based on this stage (Miyajima and Okumura 1917). The latest revision of chigger genera in the Neotropical and Nearctic regions was published in the 1970's accounting more than 100 genera, with 19 of these taxa documented for Brazil (Brennan and Goff 1977).

In the Neotropical region, the genus *Quadrasetas* Brennan, 1970 is known only from larval stage and consists in 14 ectoparasitic species associated with small-mammals; however one species, *Quadrasetas flochi* (Brennan and Jones, 1960), can also parasitize birds. Of these Neotropical representatives, *Quadrasetas brasiliensis* Goff and Gettinger, 1989 and *Quadrasetas pazca* (Brennan and Jones, 1964) have been reported from Brazil.

*Quadrasetas brasiliensis* was described from larvae collected on the rodent *Hylaeamys megacephalus* (Fisher), cited as *Oryzomys capito* (Olfers), and from the marsupials *Gracilinanus agilis* (Burmeister), cited as *Marmosa agilis*, and *Monodelphis americana* (Müller), all captured at an average of 25 km from Brasília (Federal District). In the original description of *Q. brasiliensis*, Goff and Gettinger (1989) designated the holotype based on larva collected from *H. megacephalus*, deposited in the MZUSP - Museu de Zoologia da Universidade de São Paulo, São Paulo, Brazil, and paratypes deposited in following three collections: BPBM - Bernice Pauahi Bishop Museum, Honolulu, Hawaii, USA (currently housed in the NMNH collection), OMNH - Sam Noble Oklahoma Museum of Natural History, Norman, Oklahoma, USA, and NMNH (earlier acronym USNM) - United States National Museum of Natural History, Smithsonian Institution, at United States Department of

Agriculture, Beltsville, Maryland, USA. Unfortunately no type specimens of *Q. brasiliensis* were found while examining these collections, however, we found specimens mounted on slides, collected from the same host species and locality, but different date, that allowed us to confirm the identity of our material. Here we redescribe the larva, describe the deutonymph instar obtained from field-collected larvae, report new hosts and localities, and include genetic data for this species in Brazil.

### 3.2 MATERIAL AND METHODS

#### 3.2.1 Morphological study

Slide-mounted larvae of *Quadrasetia* deposited in the NMNH, which are housed at the Systematic Entomology Laboratory (BARC-USDA-ARS), and in the Acari Collection from Butantan Institute, São Paulo, Brazil (IBSP), were examined and identified based on the original descriptions of the 14 known *Quadrasetia* species.

Measurements and drawings were made using a Leica DFC 500 digital camera coupled to a Leica DM4000B optical microscope. Extended focal range images were composed using the Leica Application Suite version 2.5.0. Optical micrographs were prepared using Adobe Photoshop v. 13.0 and Inkscape V. 2. All measurements were made in micrometers ( $\mu\text{m}$ ), followed by maximum, minimum, mean and standard deviation (SD). Micrographs of larvae and deutonymphs were made by the Low Temperature Scanning Electron Microscopy (LT-SEM) at the US Department of Agriculture, Electron and Confocal Microscopy Unit, Beltsville, Maryland as described in Dowling et al. (2010) and Bolton et al. (2014).

We followed the terminology of Goff et al. (1982), with adaptations proposed by Stekolnikov (2008) and Stekolnikov and Daniel (2012) concerning general nomenclature of larval stages. To standardize the nomenclature of the specialized setae of the Prostigmata group, we adopted the terminology used by Wohltmann (2006, 2007) and Kethley 1990. For the deutonymphs, we used the terminology proposed by Audy (1954) and Crossley (1960).

Chigger larvae collected on rodents and marsupials from the municipality of Morro Grande, Cotia, São Paulo were reared until deutonymph stage under laboratory conditions. To achieve this, part of these larvae were placed in Petri dishes, with a mixture of plaster of Paris and activated charcoal with different proportions (3:1, 2:1 and 1:1). The Petri dishes were kept in darkness at room temperature (range  $26^\circ\text{C} \pm 1^\circ\text{C}$ ), and the substrate was daily

moistened with distilled water to provide ( $98\% \pm 1\%$ ) humidity. Once deutonymphs emerged, corresponding skins left on the dish surface were mounted on slides in Hoyer's medium for identification following Walter and Krantz (2009). Eggs of *Onychiurus* sp. (Insecta: Collembola) and *Aedes aegypti* (Insecta: Diptera) were offered as food (Lipovsky 1954). One part of the field-collected larvae and laboratory-reared deutonymphs was preserved in 100% ethanol for molecular analyses. Old larval skins and the rest of the material were deposited as voucher at the IBSP collection.

### 3.2.2 Molecular analysis

DNA extraction using the Guanidine Isothiocyanate lysis protocol (Chomkzynski 1993) was individually performed to 19 larvae (IBSP12557B) collected from *Akodon montensis* and to 5 deutonymphs reared from larvae hosted by the same rodent. Each mite was placed in an Eppendorf microtube, and punctured in the idiosomal region with a sterile needle (1.20 \* 40 – 18G). After the DNA extraction all exuviae were recovered and mounted in slides, as a voucher.

A PCR targeting a  $\approx$ 500-pb fragment of the 18S ribosomal RNA gene was performed using primers Mite18S-1F (3'-ATATTGGAGGGCAAGTCTGG-5') and Mite18S-1R (3'-TGGCATCGTTATGGTAG-5') as described by Otto and Wilson (2001). For the gene COI we used the following primers targeting a  $\approx$ 400-600-pb: bcdF01 (CATTTCCHACTAACATARGATATTGG) and bcdR04 (TATAAACYTCGGATGNCCAAAAAA) as described by Dabert (2008, 2010) with adaptations of Moniuszko et al. (2015); and 772 (TGATTTTTGGTCACCCAGAAG) and 773 (TACAGCTCCTATAGATAAAC) as described by Navajas (1994). For each reaction, negative (Milli-Q water free of DNA) and positive controls (chigger mite pools) were used. All PCRs were performed in a Mastercycler Gradient (Eppendorf® California, USA). PCR products with concentrations higher than 20 ng/ $\mu$ l, were selected and purified with ExoSap-IT (GE Healthcare Pittsburgh, PA). Sanger sequencing reactions of the samples were performed in the “Centro de pesquisa sobre Genoma Humano e Células Tronco do Instituto de Biociências da USP”. Obtained sequences were assembled with Sequencing Analysis 5.3.1 software, and submitted to BLAST analyses (Altschul et al. 1990) in order to infer similarities with other mites available in GenBank. Different haplotypes were visually discriminated after an alignment using CLUSTAL W algorithm (Thompson et al. 1994) implemented in Geneious R9 (Kearse et al. 2012).

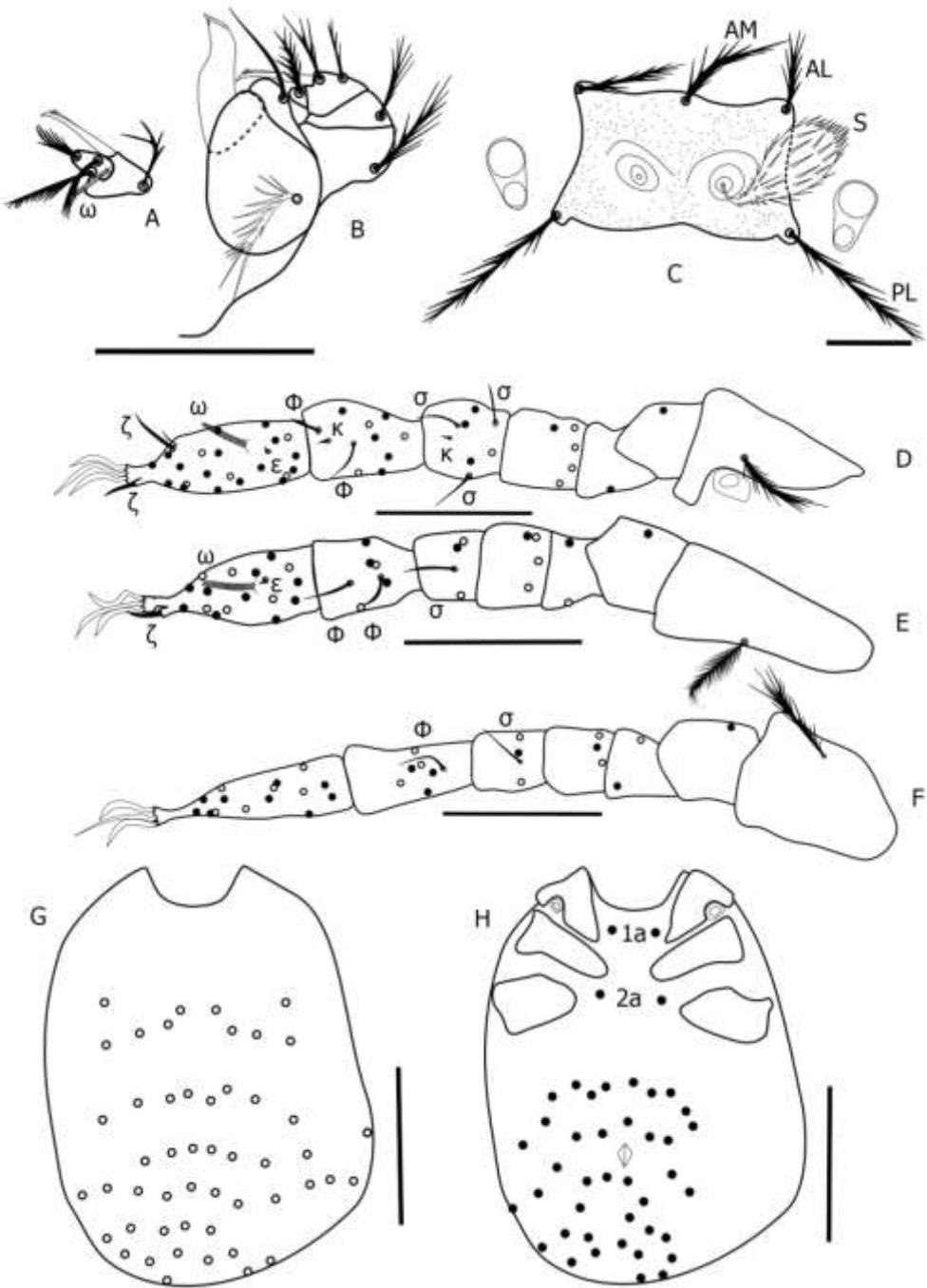
### 3.3 RESULTS

#### ***Quadraseta brasiliensis* Goff and Gettinger, 1989**

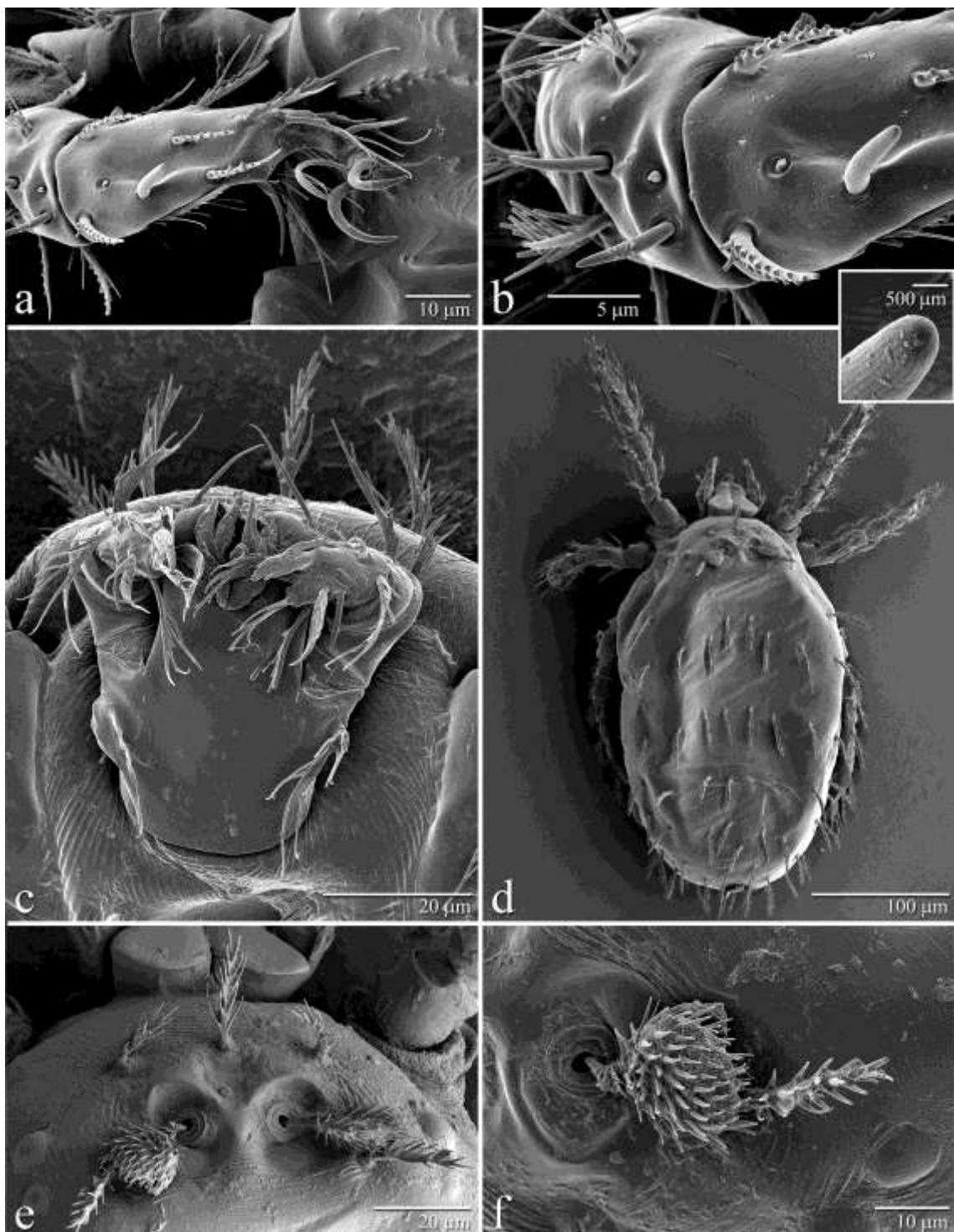
*Quadraseta brasiliensis* Goff and Gettinger, 1989: 557.

**Diagnosis.** Larva. (Figures 1, 2 and 3A; Table 1) Palpal tibia setae branched, galeal setae nude, claviform trichobothria, 2 humeral setae, the first row of idiosomal dorsal setae are arranged in line with 8 setae, genu of leg I with 3 solenidia, the base of microseta is proximal to the base of solenidion on the tarsus of leg I.

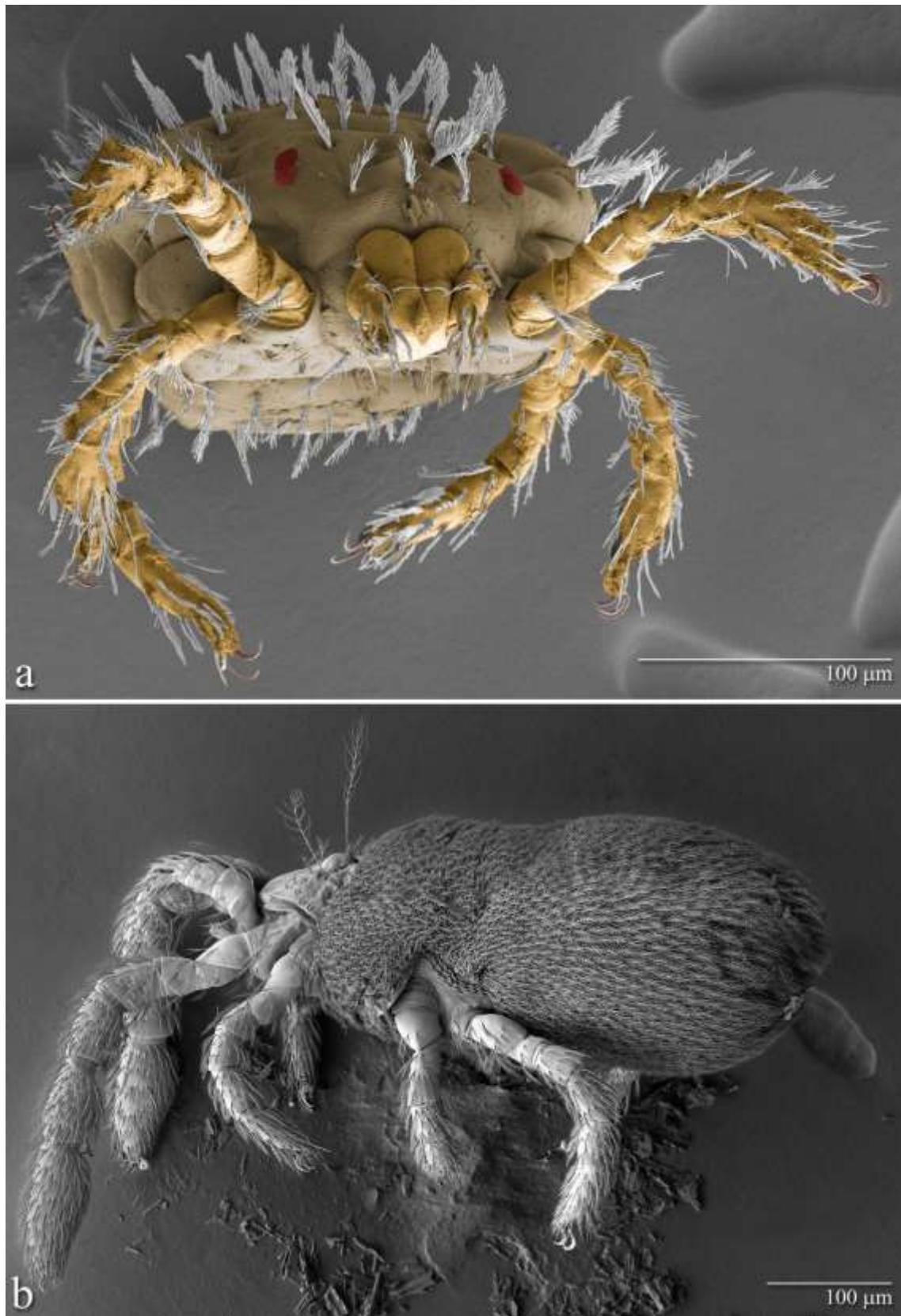
Deutonymph. (Figures 4 and 5, Table 2) Sternal area with five branched setae on one side and four branched setae on the other side between coxae I; 8 distinct but minute dorsal teeth in cheliceral each blade, palp tibia with 5 setae branched and 1 dorsal nude seta; base of tibial claw flanked by 2 spoon-shaped accessory setae, palp tarsus with 7 branched setae, 2 nude apical sensory setae, a leaf-shaped tectum, with a single tectal seta, parascutal setae number approximately six on either side of crista, typically arranged at 2-4; three pairs of branched epivalval setae and six pairs of branched centrovalval setae.



**Figure 1.** Morphological details of the neotype of *Quadraseta brasiliensis*. A. ventral view of palpal tarsus; B. dorsal view of gnathosoma; C. scutum; D. Leg I; E. Leg II; F. Leg III; G. dorsal view of idiosoma; H. ventral view of idiosoma. Black spots = ventral setae of the idiosoma; white spots = dorsal setae of the idiosoma. Symbols:  $\omega$  = solenidion of palpal tarsus; AL = anterolateral seta on scutum; PL = posterolateral seta on scutum; AM = anteromedial seta on scutum; S = trichobothria;  $\sigma$  = solenidion of the genu I, II and III;  $\kappa$  = microsetae on genu and tibia;  $\Phi$  = solenidion of the tibia I, II and III;  $\omega$  = solenidion I and II;  $\varepsilon$  = famulus I and II;  $\zeta$  = dorsal eupathid I, subterminal eupathid of the legs I and II; z = companion seta of dorsal eupathid on the leg I; 1a = anterior sternal setae; 3a = posterior sternal setae. Scales: A-F 50  $\mu$ m; G-H 100  $\mu$ m.



**Figure 2.** Scanning electron micrographs of *Quadraseta brasiliensis* larvae. A. Tarsus of the leg I; B. details of the specialized setae of tibia and tarsus of the leg I (in the small box striations of the solenidion of the tarsus of the leg I); C. ventral view of the gnathosoma; D. idiosomal dorsum; E. scutum; F. details of the trichobothria.



**Figure 3.** Low-temperature scanning electron micrographs of *Quadraseta brasiliensis*. A. frontal view of larva; B. lateral view of deutonymph.

**Redescription.** Larva. Yellowish-white colored when alive. Gnathosoma (Figure 1A and B, 2C) - palpal setal formula B/B/BBB/4B, B = branched seta on the femur; B = branched seta on genu; BBB = branched dorsal, ventral setae, and lateral tibial setae, respectively 4B = four branched with  $\omega$  on tarsus; odontus trifurcate; cheliceral blade with tricuspid cap; gnathobase punctate, galeal nude setae. Idiosoma – eyes 2/2; anterior larger, on ocular plate, *scutum* with 1 pair of AL, 1 pair of PL and a single AM seta, PL > AM > AL; 1 pair of claviform trichobothria, covered with setules; scutum with concave lateral margins, posterior margin with two smooth evaginations and anterior margin with two invaginations both turned to the base of trichobothria, as figure 1C, 2E and F. Ninety-four idiosomal setae (material range 84-97 setae), including 1 pair of humeral, 1<sup>st</sup> row of dorsal idiosomal setae with 8 setae, 2<sup>nd</sup> row with 8 setae, 3<sup>rd</sup> row with 8 setae, 4<sup>th</sup> row with 11 setae and 5<sup>th</sup> row with 13 setae, totaling 48 dorsal setae (material range 39-48 setae), 2 pairs of sternal, and 26 preanals and 14 postanals, totalizing 40 ventral setae (material range 40-47 setae) (Figures 1H and G, 2D). Legs - seven-segmented, femur divided in basifemur and telofemur, with a pair of claws and a clawlike empodium, onychotriches absent, coxae not striate. *Leg I* - coxa with 1 branched seta (1B); trochanter 1B; basifemur 1B; telofemur 5B; genu 4B,  $\sigma$  3, with  $\kappa$ ; tibia 8B,  $\Phi$  2, with  $\kappa$ ; tarsus 21B, with  $\omega$ ,  $\epsilon$ , dorsal eupathid ( $\zeta$ ) with a companion seta (z) and subterminal eupathid ( $\zeta$ ), base of  $\epsilon$  proximal to base of  $\omega$  (Figure 1D, 2A and B). *Leg II* - coxa 1B; trochanter 1B; basifemur 2B; telofemur 4B; genu 3B,  $\sigma$ ; tibia 6B,  $\Phi$  2; tarsus 16B, with  $\omega$ ,  $\epsilon$ , and subterminal eupathid, base of  $\epsilon$  proximal to base of  $\omega$  (Figure 1E). *Leg III* – coxa 1B on anterior margin, trochanter 1B; basifemur 2B; telofemur 3B; genu 3B,  $\sigma$ , tibia 6B,  $\Phi$ ; tarsus 15B (Figure 1F). The morphometric data are shown in Table 1.

**Table 1.** Measurement of *Quadrassetas brasiliensis* (Larvae)

	<b>AW</b>	<b>PW</b>	<b>SB</b>	<b>ASB</b>	<b>PSB</b>	<b>SD</b>	<b>AP</b>	<b>AM</b>	<b>AL</b>	<b>PL</b>	<b>S</b>
<b>Holotype</b> <b>(Goff and Gettinger 1989)</b>	53	69	25	26	17	43	33	36	27	45	38
<b>Paratypes</b> <b>(Goff and Gettinger 1989)</b>	51-55	63-70	23-26	21-26	15-17	-	30-33	34-40	21-27	39-46	36-39
<b>Minimum*</b>	42	49	19	19	10	28	28	21	19	31	26
<b>Maximum*</b>	49	63	23	23	12	33	34	32	32	38	36
<b>Mean*</b>	45	57	22	21	11	32	31	27	24	35	31
<b>SD*</b>	2,1	4,0	1,4	1,2	0,7	1,6	1,5	3,9	3,6	2,1	3,1

**Table 1.** (continued)

	<b>H</b>	<b>Dmin</b>	<b>Dmax</b>	<b>Vmin</b>	<b>Vmax</b>	<b>I</b>	<b>II</b>	<b>III</b>	<b>Ip</b>	<b>TaIII</b>	<b>TaW</b>	<b>ω I</b>	<b>ω II</b>	<b>length</b>	<b>width</b>	<b>1a</b>	<b>3a</b>
<b>Holotype</b> <b>(Goff and Gettinger 1989)</b>	51-55	37	44	18	35	249-262	215-224	254-259	718-745	72	16	15-16	15	550	390	40-48	29-33
<b>Paratypes</b> <b>(Goff and Gettinger 1989)</b>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<b>Minimum*</b>	32	21	40	15	32	217	181	204	610	46	13	13	14	225	165	25	23
<b>Maximum*</b>	45	31	44	17	33	252	213	252	693	62	16	14	16	632	509	42	35
<b>Mean*</b>	41	25	41	16	33	232	196	219	647	55	15	14	15	401	320	33	28
<b>SD*</b>	2,7	2,8	1,5	0,7	0,5	10,6	9,1	10,5	27,1	4,5	0,8	0,5	0,7	122,6	101,0	4,6	2,7

**Legend:** AW = distance between AL setae on scutum; PW = distance between AL setae on scutum; SB = distance between sensillary bases; ASB = distance from sensillary bases to extreme anterior margin of scutum; PSB = distance from sensillary bases to extreme posterior margin of scutum; SD = ASB + PSB; AP = distance from anterolateral setal base to posterolateral setal base on one side; AM = anteromedial seta on scutum; AL = anterolateral setae on scutum; PL = posterolateral setae on scutum; S = trichobothria; H = humeral setae; D = minimum and maximum length of dorsal idiosomal setae; V = minimum and maximum length of ventral idiosomal setae; I = length of Leg I; II = length of Leg I; III = length of Leg I; Ip = sum of leg lengths; TaIII = length of tarsus I; TaW = width of tarsus I; ω I = length of solenidion of the tarsus on leg I; ω II = length of solenidion of the tarsus on leg II; 1a = anterior sternal setae; 3a = posterior sternal setae. \* n = 50.

**Table 2.** Measurement of *Quadrassetas brasiliensis* (Deutonymph)

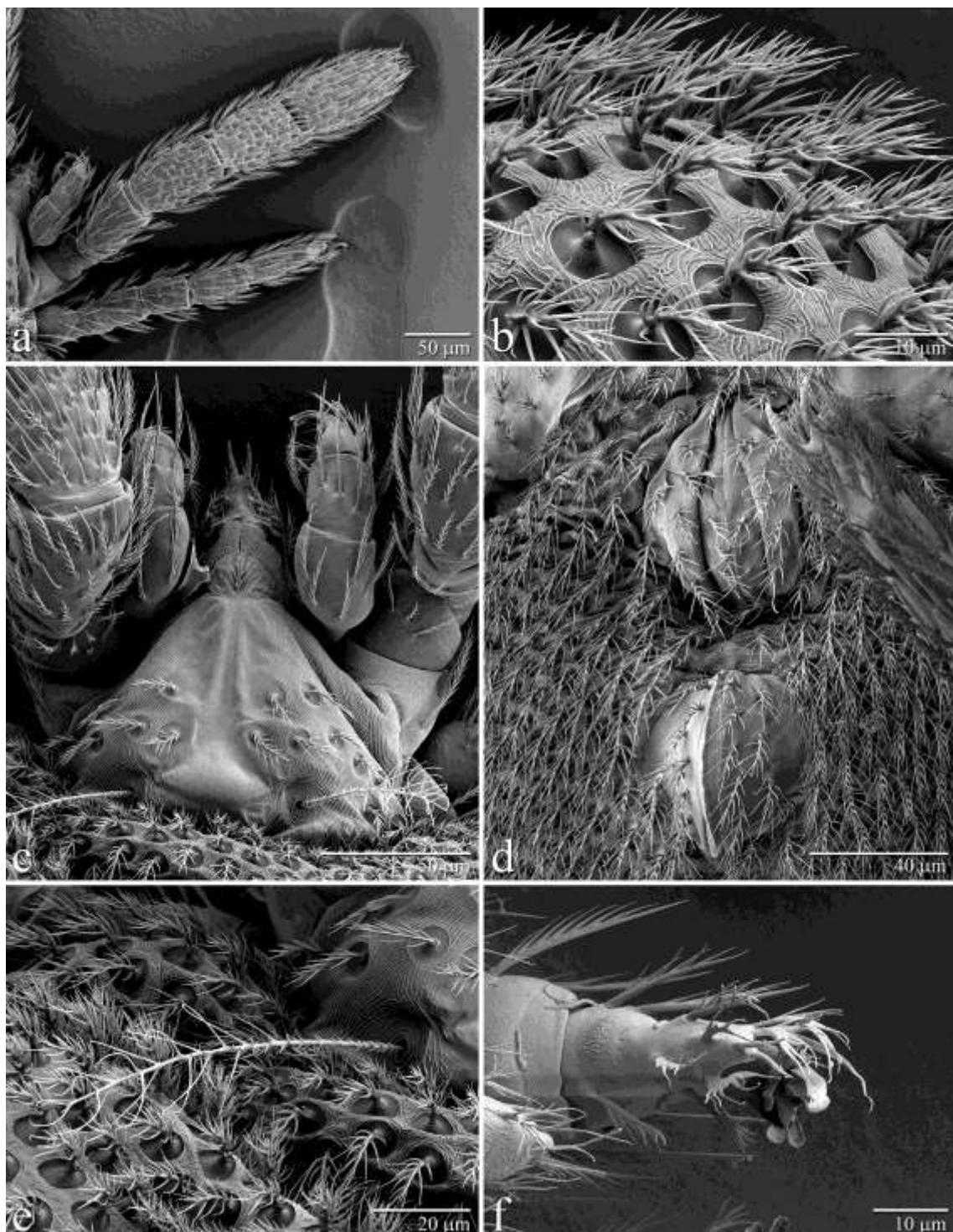
	IL	IW	ASL	PSL	CTL	SB	SENS	SS	TS	I	II	III	IV	IP	TL	TH	tL	tH	TL'	TH'	tL'	tH'	CL	BL	BH
<b>Minimum</b>	375	203	24	32	93	53	118	16	11	420	272	250	278	720	106	57	63	51	70	22	34	30	34	26	26
<b>Maximum</b>	593	357	103	82	135	64	132	27	18	502	321	395	405	1532	126	69	86	66	118	64	83	63	40	74	81
<b>Mean</b>	490	290	39	68	107	58	122	22	15	464	299	310	354	1374	117	60	76	57	78	31	45	36	39	51	49
<b>SD</b>	69,8	57,8	26,4	16,0	12,9	3,4	4,6	2,4	1,9	21,8	15,3	41,2	41,1	229,9	6,0	3,5	6,2	4,6	13,3	11,4	13,4	9,0	2,0	20,1	24,7

**Legend:** IL: total length of the idiosoma; IW: total width of the lidiosoma; ASL: distance from the base of the tectal base to the sensillary bases; PSL: distance from the sensillary bases to the end of the posterior border; CTL: distance from the base of the tectal base to the end of the posterior border; SB: distance between sensillary bases; SENS: lenght of the trichobothria; SS: length of parascutal setae; TS: length of tectal seta; I = length of Leg I; II = length of Leg I; III = length of Leg I; IV = length of Leg I; Ip = sum of leg lengths; TL: length of tarsus I; TH: width of tarsus I; TL: length of tibia I; TH: width of tibia I; TL': length of tarsus II; TH': width of tarsus II; TL': length of the tibia II; TH': width of tibia II; CL: length of the cheliceral blade; BL: length of the chelicera base (chelobase); BH: width of chelicera base (chelobase).

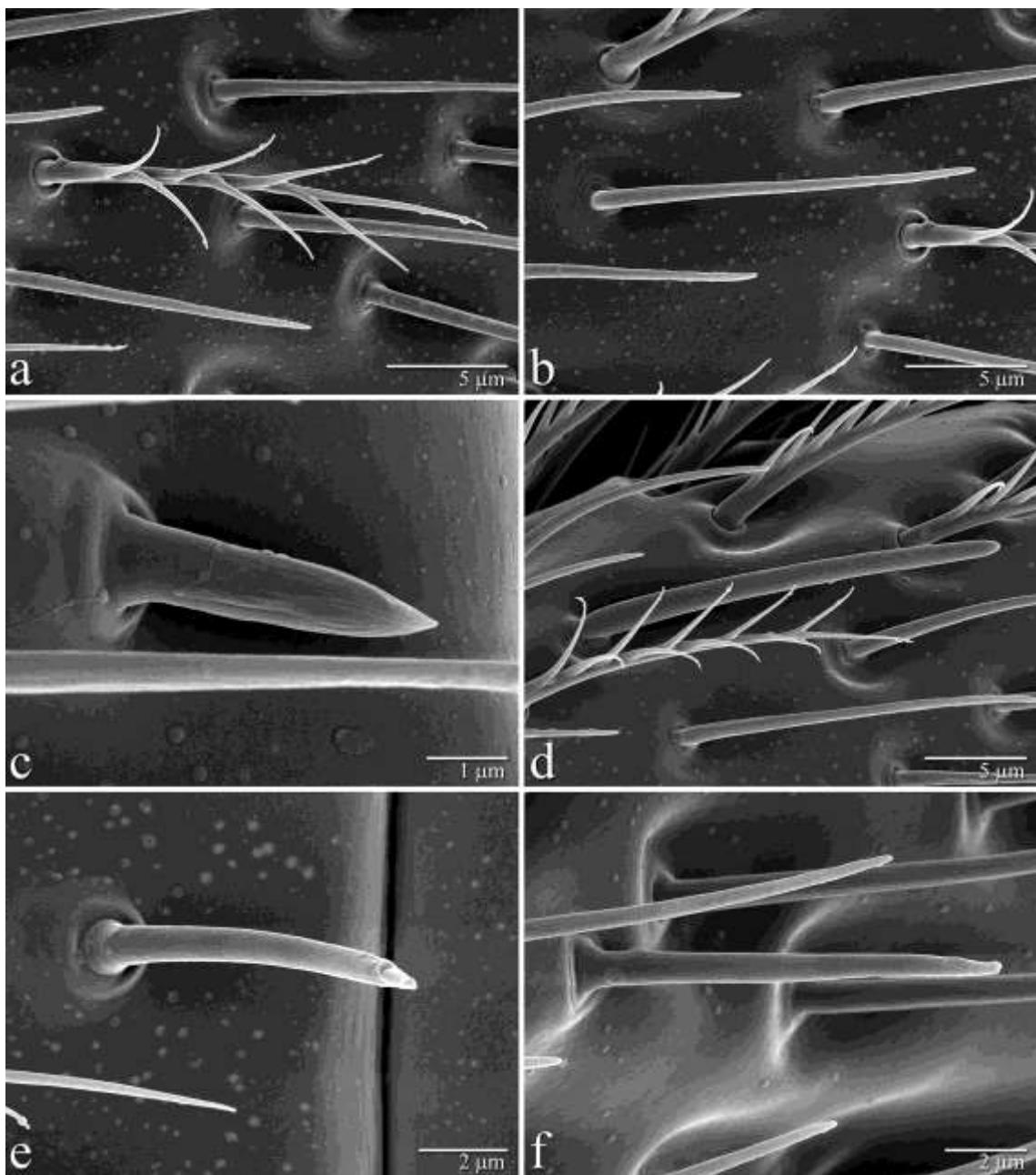
**Description.** Deutonymph. Dorsal idiosoma - color whitish yellow, eyes absent; The entire body with branched setae on plates, shown in Figure 4B; a leaf-shaped tectum, with a single tectal seta; six parascutal setae on either side of crista, typically arranged at 2-4 (Figure 4C); trichobothria long and slender, proximal portion with numerous short branches, and the distal portion with numerous long thin branches (Figure 4C and E). Ventral idiosoma - Sternal area: with five branched setae on one side and four branched setae on the other side between coxae; six pairs of branched epivalval setae, 2 pairs of oval acetabula present, posterior acetabulum smaller than anterior and three pairs of branched centrovalval setae (Figure 4D); a pair of anal plates, each one with eight branched setae (Figure 4D). Gnathosoma - Cheliceral blade straight, V-shaped, with 8 distinct but minute dorsal teeth, along each. Entire hypostome not divided into lobes, with 4 pairs of nude apical setae; palp femur with 8 branched setae; palp genu with 7 branched setae; palp tibia with 5 setae branched and 1 dorsal nude seta; base of tibial claw flanked by 2 spoon-shaped accessory setae; palp tarsus with 8 branched setae, 2 nude apical sensory setae ( $\zeta$ ) and 1 basal sensory rod ( $\omega$ ) (Figure 4F). Legs - seven-segmented, femur divided in basifemur and telofemur, tarsi with a pair of claws, onychotriches absent; leg I and IV longer than legs II and III; tarsus I oblong in shape, inflated, always longer than tibia I (Figure 4A). All leg segments densely covered with normal setulated setae and the specialized setae present on leg I (tFe-Ta), II (tFe-Ta), III (tFe-Ta), IV (Ge-Ta) (Figure 5A-F). The morphometric data are shown in Table 2.

**Remarks.** The species *Q. brasiliensis* resembles the species *Q. pazca* and *Q. mackenziei* (Yunker and Brennan, 1964), but it has the first row of dorsal idiosomal setae in lines and the all palpal tibia setae branched. The species *Q. brasiliensis* differs from the other species in presents 8 setae in the first row of idiosomal dorsal setae, while the other two species have 10 setae. *Q. brasiliensis* and *Q. mackenziei* has galeal nude setae, while *Q. pazca* has galeal branched setae. The trichobothria in *Q. brasiliensis* and *Q. pazca* are clavate, while in *Q. mackenziei* are globose.

**Biology.** In laboratory conditions, engorged larvae, recently collected from the hosts, kept in Petri dishes with substrate (1:1), remained active for 1-3 days. After this period the larvae enter in the process of quiescence (protonymph), at this stage there is an extrusion of the legs. This process lasted 7-11 days and a deutonymph emerged from the anterior dorsal portion of protonymph, the deutonymphs remained alive for up to 20 days without feeding.



**Figure 4.** Low-temperature scanning electron micrographs of *Quadraseta brasiliensis* deutonymph. a. general view of the palp, leg I and II; b. idiosomal dorsal setae; c. crista metopica; d. genital area and anal plates; e. trichobrothrium; f. palpal tarsus and palpal tibia.



**Figure 5.** Low-temperature scanning electron micrographs of *Quadraseta brasiliensis* deutonymph. Details of the deutonymphs' setae. a. branched setae of the genu on the leg I; b. solenidion of the genu on the leg I; c. microseta of the genu on the leg I; d. solenidion of the genu on the leg I; e. solenidion of the femur on the leg I; f. solenidion of the tibia on the leg I. Scales: a, b and d 5  $\mu\text{m}$ ; c 1  $\mu\text{m}$ ; e and f 2  $\mu\text{m}$ .

**New records (Figure 6)** BRAZIL – 6 larvae, Fazenda Água Limpa (FAL), Brasilia - Federal District, (15°56'45"S, 47°56'8"W), 21-V-1984, *H. megacephalus* (DG494); 10 larvae, same locality, 24-V-1984, *Monodelphis americana* (DG498); 10 larvae, same locality and host (DG515), 16-VI-1984; 10 larvae, same locality, 21-II-1984, *Monodelphis* sp. (DG260); 10 larvae, same locality, 22-IV-1984, *H. megacephalus* (DG394); 10, same locality, 24-II-1984,

*H. megacephalus* (#102); 10 larvae, Reserva Ecológica do Instituto Brasileiro de Geografia e Estatística (IBGE) ( $15^{\circ} 56'41"S, 47^{\circ} 53'07"W$ ), 8-VII-1984, no host information (#37); 1 larva (IBSP 1154), Serra da Cantareira, São Paulo, state of São Paulo ( $23^{\circ} 22'44"S, 46^{\circ} 31'38"W$ ), 8-IX-1937, *Cuniculus paca*, Navas J. coll.; 8 larvae (IBSP 10390), Adrianópolis, state of Paraná ( $24^{\circ} 39'25"S, 48^{\circ} 59'27"W$ ) 12-IX-2009, *Euryoryzomys russatus*; 1 larva (IBSP10521A), Zoológico de São Paulo, São Paulo, state of São Paulo ( $23^{\circ} 38'57.49"S, 46^{\circ} 37'19.76"W$ ), 16-IV-2010, *Akodon* sp.; 1 larva (IBSP 10522), Barra do Una, Peruíbe, state of São Paulo ( $24^{\circ} 19'12"S, 46^{\circ} 59'52"W$ ), 8-XI-2010, *Euryoryzomys russatus*; 2 larvae (IBSP 10524A), same locality and host, 18-VI-2010; 1 larva (IBSP 10525A), same data; 2 larvae (IBSP 10526A), same locality and host, 16-VI-2010; 4 larvae (IBSP 10527A), same data; 1 larva (IBSP 10528), same locality and host, 7-XI-2010; 2 larvae (IBSP 10529), same locality and host, 21-IV-2010; 2 larvae (IBSP 10530), same locality and host, 20-IV-2010; 2 larvae (IBSP 10531), same locality and host, 21-IV-2010; 3 larvae (IBSP 10532), same data; 3 larvae (IBSP 10533A), same locality and host, 19-IV-2010; 5 larvae (IBSP 10535A), Serra da Cantareira, São Paulo, state of São Paulo, same host, 12-IV-2011; 2 larvae (IBSP10536), same locality and host, 14-IV-2011; 10 larvae (IBSP 10537), same data; 3 larvae (IBSP10538), same locality and host, 18-XII-2010; 4 larvae (IBSP 10540), same locality and host, 17-II-2010; 1 larva (IBSP 10590D), Condomínio Vila Verde, Itapevi, state of São Paulo, ( $23^{\circ} 32'56"S, 46^{\circ} 56'02"W$ ), 12to16-XII-2011, *Didelphis aurita*; 6 larvae (IBSP 11096G), Morro Grande, Cotia, state of São Paulo ( $23^{\circ} 38'58.12", 46^{\circ} 57'45.99"W$ ), 20-VI-2012, *Euryoryzomys russatus*; 9 larvae (IBSP 11116B), Condomínio Vila Verde, Itapevi, state of São Paulo, 29-XI-2012, *Monodelphis* sp.; 1 larva (IBSP 11130), Morro Grande, Cotia, state of São Paulo, 22-VII-2012, *Monodelphis* sp.; 2 larvae (IBSP11131), same locality, 19-X-2012, *Euryoryzomys russatus*; 1 larva (IBSP 11132), same locality, 23-VII-2012, *Thaptomys nigrita*; 1 larva (IBSP 11178C), same locality, *Akodon montensis*; 4 larvae (IBSP 11259C), same locality, 18-IV-2013, *Euryoryzomys russatus*; 2 larvae (IBSP 11303C), Sete Barras, state of São Paulo ( $24^{\circ} 23'16"S, 47^{\circ} 55'33"W$ ), 20-IV-2013, *Olygoryzomys nigripes*; 2 larvae (IBSP 11304A), same locality, 20-IV-2013, *Olygoryzomys* sp.; 2 larvae (IBSP 11305), same locality, 10-VIII-2013, *Euryoryzomys russatus*; 1 larva (IBSP 11365), same data; 4 larvae (IBSP 11366), same locality, 10-VIII-2013, *Monodelphis iheringi*; 2 larvae (IBSP 11367), same locality, 15-VIII-2013, *Oxymycterus* sp.; 2 larvae (IBSP 11369), same locality, no date, *Delomys* sp.; 1 skin of larva and 3 deutonymphs (IBSP 12138B), Morro Grande, Cotia, state of São Paulo, no date, *Euryoryzomys russatus*; 1 skin of larva and 3 deutonymphs (IBSP 12140), same locality, no date, *Akodon* sp.; 1 deutonymph (IBSP 12141), same locality, no

date and host information; 1 skin of larva (IBSP 12144), same locality, 15-IV-2015, *Akodon* sp.; 3 skins of larva and 3 deutonymphs (IBSP 12146A), same locality and date, *Monodelphis* sp.; 2 skins of larva and 2 deutonymphs (IBSP 12554A), same locality, no date and host information; 27 larvae, 15 skins of larva and 11 deutonymphs (IBSP 12557B), same locality, 20-VIII-2015, *Akodon montensis*; 4 skins of larva (IBSP 12559), same locality, no date and host information; 12 skins of larva (IBSP 12560), same locality, no date and host information; 1 skin of larva (IBSP 12695), 23-X-2013, *Thaptomys nigrita*.



**Figure 6.** Geographical distribution of *Quadraseta brasiliensis*.

**Molecular analysis.** Although expected size amplicons were obtained for 14/24 of the samples submitted to PCR, only three larvae and two deutonymphs yielded bands strong enough for sequencing. After an alignment of 421-bp including these five sequences, two haplotypes differing in one nucleotide were observed: one belonging to the deutonymphs and two larvae (GenBank accession numbers: KY934462, KY934464, MF113412 and MF113413), and another to one larva (KY934463). By BLAST analyses, both haplotypes of *Q. brasiliensis* were 96 – 99% similar to the sequences available in GenBank for *Allothrombium* sp. (KP276493) and *Eutrombicula splendens* (Ewing, 1913) (KP325057) respectively. For the gene COI, none of the tested samples amplified.

### 3.4 DISCUSSION

According to the original description of this species by Goff and Gettinger (1989), the holotype and paratypes of *Q. brasiliensis* were deposited in the BPBM, MZUSP, OMNH and NMNH collections. However, a search of these collections revealed no type specimens. Nevertheless, in the NMNH we found additional slide-mounted material coinciding with the same data of collection for the type specimens, except for the collecting date. After a review of all of the additional material, we found variation only in the position of the dorsal, preanal and postanal setae, depending on the degree of engorgement of the mites, as well as in the total number of idiosomal setae.

We have succeeded in obtaining deutonymphs, by rearing larvae on activated charcoal as substrate in order to remove plaster of Paris toxicity (Shatrov 2003). A great variety of substrate types have been tested successfully by other researchers (Wharton 1946; Wharton and Carver 1946; Michener 1946; Jenkins 1947; Farrel and Wharton 1949; Hyland 1951, 1961; Lipovsky 1953, 1954; Kaufman and Traub 1966; Nadchatram 1968; Everett et al. 1973; Mallow and Crossley 1984; Takahashi et al. 1993; Shatrov 1996; Tuegel and Wrenn 1998), including modifications in the proportion of activated charcoal, density of those substrates and sizes or kind of the culture containers.

In the present study, the deutonymphs of *Q. brasiliensis* were obtained under temperatures of 26 °C ( $\pm 1$  °C), as well as the substrate with activated charcoal (1:1) demonstrated to be more efficient than in other proportion. Thus, the following parameters proved to be determinants of successful acquisition of deutonymph stages of *Q. brasiliensis* in laboratory conditions. The relative humidity inside the dish must be near the saturation level without being completely saturated in order to avoid the formation of fungi colonies that would need to be removed immediately.

Lipovsky (1951) tested different types of food, such as eggs of Lepidoptera, Coleoptera, Collembola, and Diptera for *Eutrombicula alfreddugesi* (Oudemans, 1910) and *Eutrombicula splendens* (Ewing, 1913), noticing that the preferred food source varies depending of the trombiculid species. Specimens of *T. alfreddugesi* and *T. splendens* fed on eggs Collembola as well as of Diptera; whereas *Hirsutiella zachvatkini* (Schluger, 1948) (Shatrov 2003), *Eutrombicula cinnabaris* (Ewing, 1920) (Tuegel and Wrenn 1998), *Neoschongastia americana* (Hirst, 1921) (Everett et al. 1973), *Leptotrombidium pallidum* (Nagayo, Miyagawa, Mitamura and Tamiya, 1919) (Takahashi et al. 1993), and *Hannemania hegneri* Hyland, 1956 (Hyland 1961) fed almost exclusively on *Sinella curviseta*

(Collembola) eggs, in laboratory rearing. Eggs of *Onychiurus* sp. (Collembola) were given daily (at least 10 eggs per day) presented pink-like color (by fungi infection), and this fact was previously recorded by Lipovsky (1951). This author verified that eggs were infected with *Penicillium*, caused progressive lethargy which led to death of the specimens of *E. alfreddugesi* and *E. splendens*.

Temperature control was not a high priority issue for colony maintenance of *Eutrombicula batatas* (Linnaeus, 1758) and *Blankaartia alleei* (Ewing, 1926), since temperature variations of  $\pm 5^{\circ}\text{C}$  (Jenkins, 1947) did not affect the success of the colony. Shatrov (2003) observed the same situation for *H. zachvatkini*.

The only record of *Q. brasiliensis* was the type locality described from the Brasília (Federal District), collected on rodents *H. megacephalus* (cited as *Oryzomys capito*) and on marsupials *G. agilis* (cited as *Marmosa agilis*) and *M. americana* (Goff and Gettinger 1989). In this study, the species was recorded in the southern region of Adrianópolis (state of Paraná), and *E. russatus* was a new host record. We also added the cities of São Paulo, Cotia, Itapevi, Peruíbe and Sete Barras as new occurrences (Figure 5), and rodents of the species: *A. montensis*, *C. paca*, *O. nigripes* and *T. nigrita*; and marsupials of the species *D. aurita* and *M. iheringi* as new hosts for *Q. brasiliensis*.

Partial sequences obtained for the 18S ribosomal RNA gene of two larvae and two deutonymphs were identical between each other, and one larva yielded a sequence differing only in one nucleotide. BLAST analyses sequences of *Q. brasiliensis* were highly similar (99%) to *E. splendens*, the comparison of the complete sequences of both species should be done in order to draw further conclusions. These sequences correspond to the first molecular identification of the genus *Quadrasetata*.

Kampen (2004) succeeded in amplifying the COI gene of *Neotrombicula autumnalis* (Shaw, 1790) larvae using primers for Tetranychidae mites designed by Navajas (1994); moreover, Moniuszko et al. (2015) were successful in amplifying the same gene, from larvae and deutonymphs of *Hirsutiella zachvatkini* (Schluger, 1948), using the primers by Dabert et al. (2008; 2010). In the present study, both pairs of primers were tested and none of them amplified the COI gene for *Q. brasiliensis*.

This is the first description and record of *Q. brasiliensis* deutonymphs that developed from larvae maintained in laboratory conditions. The only previous description of the deutonymph instar of a chigger species recorded for Brazil was that of *Eutrombicula ralpauddyi* Vercammen-Grandjean and André, 1966.

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**4. CHAPTER 3 - DESCRIPTION OF *Parasacia fernandae* SP.N.  
(TROMBIDIFORMES: TROMBICULIDAE) AND NEW RECORDS OF  
CHIGGERS FROM RODENTS IN RIO DE JANEIRO STATE, BRAZIL**

**4.1 INTRODUCTION**

Chiggers - the larval stage of Trombiculidae - are ectoparasites of vertebrates. During the feeding process, these mites inject digestive enzymes into the skin of the hosts, which can cause severe skin reactions, commonly known as trombiculiasis. Some species of chiggers can also transmit pathogens (Ewing 1944). Jacinavicius et al. (2018b) reported 63 chigger species parasitizing vertebrates, including rodents in Brazil. In the state of Rio de Janeiro, seven chigger species have been recorded. Six of them—from birds, reptiles, primates and bats, as follows: *Blankaartia sinnamaryi* Floch et Fauran, 1956; *Eutrombicula batatas* (Linnaeus, 1758); *Fonsecia travassosi* (Fonseca, 1936); *Microtrombicula brennani* Goff, Whitaker et Dietz, 1986; *Perissopalla ipeani* Brennan, 1969; and *Speleocola tamarina* Goff, Whitaker et Dietz, 1987 (Fonseca 1936; Confalonieri and Benez 1976; Goff et al. 1986, 1987; Almeida et al. 2011; Bassini-Silva et al. 2017). The only chigger associated with rodents was *Caamembecaia gratiosus* Gazeta, Amorim, Bossi, Linhares et Serra-Freire, 2006. It was found in the Itatiaia National Park, on *Trinomys gratiosus* (Moojen, 1948) (Gazeta et al. 2006). Currently, the genus *Parasacia* Loomis, 1966 comprises 18 species that parasitize reptiles, birds and mammals in the Neotropical and the Nearctic regions (Goff 1992; Goff and Gettinger 1995; Stekolnikov and González Acuña 2015). Here, we describe a new species of this genus, and report new records of six chigger species, collected from cricetid rodents in the Parque Nacional Restinga de Jurubatiba, Macaé Municipality, Rio de Janeiro State, Brazil.

**4.2 MATERIAL AND METHODS**

The mites were slide-mounted in Hoyer's medium (Walter and Krantz 2009); they are housed at the Collection of Apterous Arthropod Vectors of Importance for Public Health (Coleção de Artrópodes Vetores Ápteros de Importância em Saúde das Comunidades), Oswaldo Cruz Institute, Rio de Janeiro, Brazil (CAVAISC—Fiocruz). The species were identified to genus using the key by Brennan and Goff (1977) and to species using the original descriptions of species in the genus. Morphological illustrations were made using Olympus BX 40 with a camera lucida. Extended focal range images were created using Leica

Application Suite v. 2.5.0. The images were prepared using Adobe Photoshop v. 13.0, and Inkscape v. 2. The terminology used in this description was adapted from Goff et al. (1982), Stekolnikov (2008) and Stekolnikov and Daniel (2012). We also used the nomenclature proposed by Kethley (1990), Wohltmann et al. (2006, 2007) and Bassini-Silva et al. (2017) for the specialized leg setae, dorsal opisthosomal setae and setae on the prodorsal sclerite.

#### 4.3 SYSTEMATICS

A total of 24 chiggers were examined and six species were identified: *Arisocerus hertigi* (Brennan et Jones, 1964); *Eutrombicula goeldii* (Oudemans, 1910); *Microtrombicula brachytrichia* Brennan, 1971; *Parasacia manueli* (Brennan and Jones, 1960); *Quadraseta brasiliensis* Goff et Gettinger, 1989; and *Serratacarus dietzi* Goff et Whitaker, 1984. We have also recorded a new species of *Parasacia* parasitizing *Nectomys squamipes* (Brants, 1827) in Parque Nacional Restinga de Jurubatiba, Macaé Municipality, Brazil. In two cases, different species of chiggers co-parasitized the same host. One case involved *P. manueli* and the new species. The other case involved *P. manueli* and *M. brachytrichia*. In both cases, the chiggers parasitized *N. squamipes*.

#### **Family Trombiculidae Ewing, 1944**

#### **Genus *Parasacia* Loomis, 1966**

**Diagnosis.** Larva. Seven branched setae on palptarsus, with  $\zeta$ ; odontus trifurcate; cheliceral blade with tricuspid cap; galealae branched or nude; scutum rectangular, with or without sinuous anterior and posterior margins, se (=PL) on the prodorsal sclerite; femora of legs I–III each divided into a basifemur and telofemur; coxae unisetose; empodium present and onychodrites absent; Leg I and II with subterminal eupathid; tarsus of the leg I with dorsal eupathid and  $\zeta$ ; mastiseta on tarsus of the leg III present or absent.

#### ***Parasacia fernandae* Jacinavicius et Bassini-Silva sp.n.**

Fig. 1 A–J, Tables 1–2

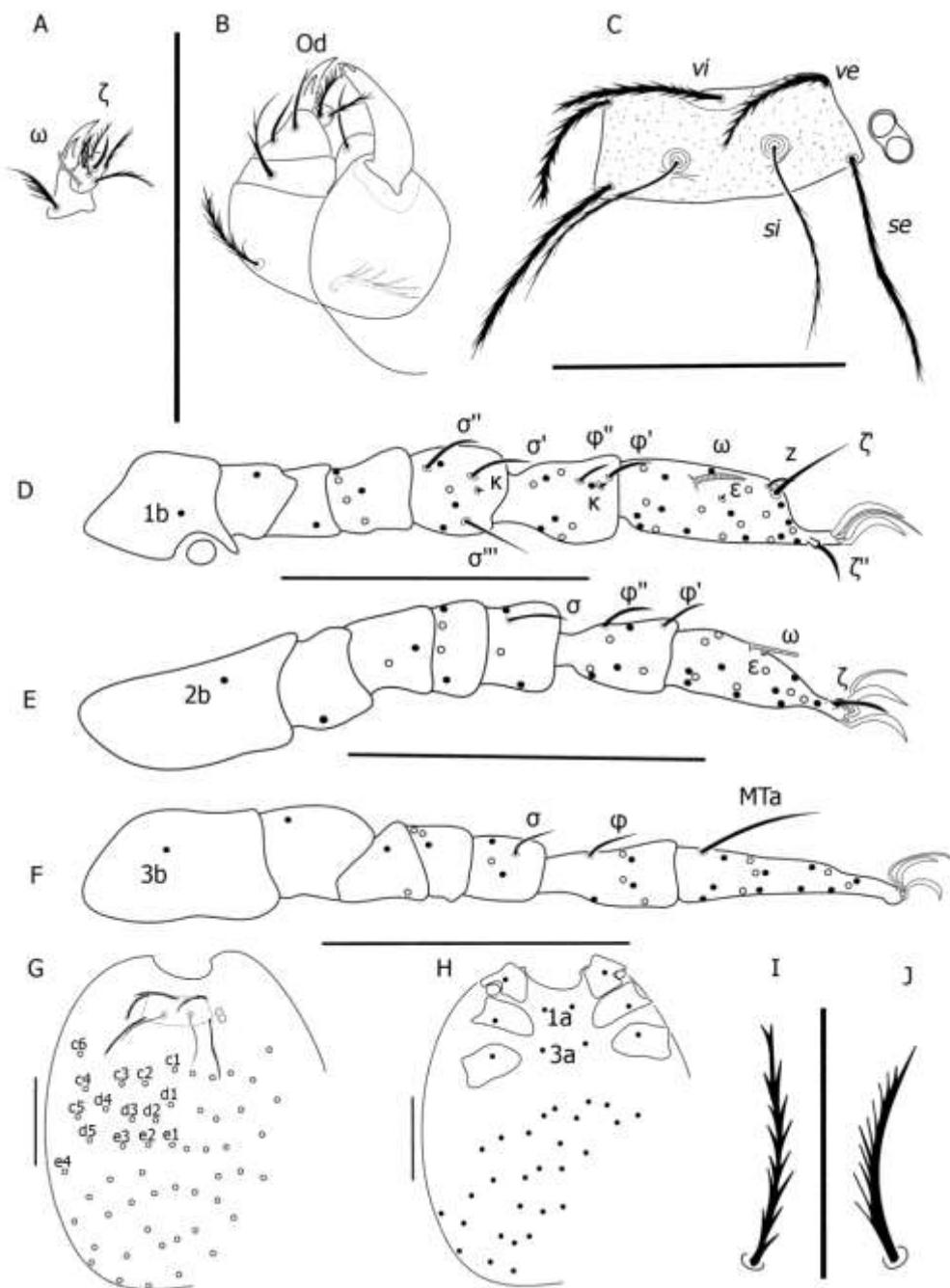
**Diagnosis.** Larva. Palptibia with nude dorsal and lateral setae, and a branched ventral seta, palptarsus with seven branched setae,  $\omega$  and  $\zeta$ , galeal setae nude, odontus trifurcate; idiosoma hypertrichous with more than 100 setae; genu of legs I each with three  $\sigma$ , tarsus of the legs I

and II each with the famulus ( $\varepsilon$ ) positioned distal to  $\omega$ , tarsus of the legs III with one mastiseta.

**Description.** Larva. Gnathosoma - palp setal formula B/B/NNB/7B $\zeta$  $\omega$ ; supracoxal setae branched, odontus trifurcate, cheliceral blade with tricuspid cap; galeal setae nude (Fig. 1A–B). Idiosoma - eyes 2/2 set in an ocular plate, anterior eye larger; prodorsal sclerite with 1 pair of flagelliform internal scapular (si) setae (trichobothria), 1 pair of vi (=AM), 1 pair of ve (=AL) seta and one pair of se on the prodorsal sclerite; se>si>vi>ve; the anterior and posterior margin of the prodorsal sclerite straight, lateral margins slightly concave (Fig. 1C). The idiosoma in all specimens are damaged, ca. 85 idiosomal setae, dorsal opisthosoma with 6 pairs of setae in the C row, with the c6 pair of seta in an anterior position (=humeral setae), D row with 5 pairs of setae, E row with 4 pairs of setae, plus ca. 23 setae, totaling ca. 53 dorsal opisthosomal setae; 2 pairs of sternal setae (1a, 3a), ca. 28 ventral setae. (Fig. 1G–J). Legs-femur legs I–III each divided into a basifemur and telofemur, each leg terminated with a pair of claws and a claw-like empodium, without onychotriches, coxal fields not striate. Leg I—coxal field with 1 branched seta 1b (1B); trochanter 1B; basifemur 1B; telofemur 5B; genu 4B, 3  $\sigma$  and  $\kappa$ ; tibia 8B, 2  $\varphi$  and  $\kappa$ ; tarsus 22B,  $\omega$ ,  $\varepsilon$ , dorsal eupathid ( $\zeta$ ) with a companion seta (z) and terminating with a subterminal eupathid ( $\zeta$ ), famulus ( $\varepsilon$ ) distal to  $\omega$  (Fig. 1D). Leg II—coxal field seta 2b (1B); trochanter 1B; basifemur 2B; telofemur 4B; genu 3B,  $\sigma$ ; tibia 6B, 2  $\varphi$ ; tarsus 15B,  $\omega$ ,  $\varepsilon$  and a subterminal eupathid ( $\zeta$ ), base of  $\varepsilon$  distal to  $\omega$  (Fig. 1E). Leg III—coxal field seta 3b (1B), trochanter 1B; basifemur 2B; telofemur 3B; genu 3B,  $\sigma$ ; tibia 6B,  $\varphi$ ; tarsus 14B with one mastiseta (Fig. 1F).

**Type material.** Holotype: larva (ACA-2625), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brazil, 15.V.2001, leg. F.M. Hatano. Paratypes: 2 larvae (ACA-2625), same data; 4 larvae (ACA-2623), same host, locality and collector, 19.V.2001; 1 larva (ACA-2618b), same host, locality and collector, 17.III.2002.

**Type deposition.** Holotype and five paratypes are deposited in CAVAISC-Fiocruz, Rio de Janeiro, Brazil; two paratypes are deposited in Acari Collection of Butantan Institute (IBSP), São Paulo, Brazil.



**Fig. 1.** Morphological details of *Parasecia fernandae* sp.n. A—ventral view of palpatarsus; B—dorsal view of gnathosoma; C—prodorsal sclerite; D—Leg I; E—Leg II; F—Leg III; G—dorsal view of idiosoma; H—ventral view of idiosoma; I—dorsal opisthosomal setae; J—ventral opisthosomal setae. Black spots—ventral setae of the idiosoma or legs; white spots—dorsal setae of the idiosoma or legs. Symbols:  $\zeta$ —eupathidium of palpatarsus and tarsus of legs I and II;  $\omega$ —solenidion of palpatarsus and tarsus of legs I and II; Od—odontus; ve—anterolateral setae; se—posterolateral setae; vi—anteromedial seta; si—trichobothria;  $\sigma$ —solenidia on the genu of legs I, II and III;  $\kappa$ —microsetae on genu and tibia of the leg I;  $\varphi$ —solenidia on the tibia of the legs I, II and III;  $\varepsilon$ —famulus on the tarsus of legs I and II; z—companion seta on tarsus of the leg I; MTa—masti setae on tarsus of the leg III; 1a—anterior sternal setae; 3a—posterior sternal setae; 1b—seta of the coxal field of the leg I; 2b—seta of the coxal field of the leg II; 3b—seta of the coxal field of the leg III. Scale bars=100  $\mu\text{m}$  (A–H), 50  $\mu\text{m}$  (I–J).

**Table 1.** Standard measurements of *Parasacia fernandae* sp. n. (n=8)

	AW	PW	SB	ASB	PSB	SD	P-PL	AP	vi	ve	se	si	H	1a	3a	DMIN	DMAX	VMIN	VMAX
<b>Holotype</b>	76	86	36	23	16	39	9	28	58	46	82	71	65	46	40	48	60	31	40
<b>Minimum</b>	74	85	33	21	15	39	8	26	55	45	79	67	62	41	37	45	54	31	35
<b>Maximum</b>	76	86	37	24	20	42	12	32	60	49	85	72	65	47	40	50	64	33	41
<b>Mean</b>	75	86	35	23	18	41	10	29	58	47	82	70	64	44	39	48	59	32	38

**Legend:** AW = distance between the bases of the *ve* setae; PW = distance between the bases of the *se* setae; SB = distance between sensillary bases; ASB = distance from sensillary bases to extreme anterior margin of the prodorsal sclerite; PSB = distance from sensillary bases to extreme posterior margin of the prodorsal sclerite; SD = ASB + PSB; P-PL = distance from *se* setal base to extreme posterior margin; AP = distance between the bases of *ve* and *se*; *vi* = anteromedial seta; *ve* = anterolateral setae; *se* = posterolateral setae; *si* = trichobothria; H= humeral setae; 1a = anterior sternal setae; 3a = posterior sternal setae; DMIN = minimum length of dorsal opisthosomal setae; DMAX = maximum length of dorsal opisthosomal setae; VMIN = minimum length of ventral idiosomal setae and VMAX = maximum length of ventral idiosomal setae.

**Table 2.** Standard measurements of the specialized setae of *Parasacia fernandae* sp. n. (n=8)

	I	II	III	Ip	$\sigma' I$	$\sigma'' I$	$\sigma''' I$	$\kappa I$	$\varphi' I$	$\varphi'' I$	$\kappa I$	$\omega I$	$\varepsilon I$	z	$\zeta' I$	$\zeta'' I$	$\sigma II$	$\varphi' II$	$\varphi'' II$	$\omega II$	$\varepsilon II$	$\zeta II$	$\sigma III$	$\varphi III$	MTa
<b>Holotype</b>	235	222	288	744	18	18	23	1	14	11	2	16	1	8	28	13	17	14	17	13	1	16	16	17	39
<b>Minimum</b>	212	219	278	698	17	17	21	1	13	11	2	16	1	8	26	13	17	14	15	13	1	15	16	17	39
<b>Maximum</b>	247	234	290	774	18	18	23	1	14	12	2	19	1	8	28	14	18	15	17	15	1	16	17	19	41
<b>Mean</b>	230	227	284	736	18	18	22	1	14	12	2	18	1	8	27	14	18	15	16	14	1	16	17	18	40

**Legend:** I = length of leg I; II = length of leg II; III = length of leg III; Ip = sum of leg lengths (coxal field to tarsus);  $\sigma' I$  = length of solenidia ( $\sigma'$ ) on genu of the leg I;  $\sigma'' I$  = length of solenidia ( $\sigma''$ ) on genu of the leg I;  $\sigma''' I$  = length of solenidia ( $\sigma'''$ ) on genu of the leg I;  $\kappa I$  = length of microseta on genu of the leg I;  $\varphi' I$  = length of solenidia ( $\varphi'$ ) on tibia of the leg I;  $\varphi'' I$  = length of solenidia ( $\varphi''$ ) on tibia of the leg I;  $\kappa I$  = length of microseta on tibia of the leg I;  $\omega I$  = length of solenidion on tarsus of the leg I;  $\varepsilon I$  = length of famulus on tarsus of the leg I; z = length of the companion seta on tarsus of the leg I;  $\zeta' I$  = length of dorsal eupathidium on tarsus of the leg I;  $\zeta'' I$  = length of subterminal eupathidium on tarsus of the leg I;  $\sigma II$  = length of solenidion on genu of the leg II;  $\varphi' II$  = length of solenidia ( $\varphi'$ ) on tibia of the leg II;  $\varphi'' II$  = length of solenidia ( $\varphi''$ ) on tibia of the leg II;  $\omega II$  = length of solenidion on tarsus of the leg II;  $\varepsilon II$  = length of famulus on tarsus of the leg II;  $\zeta II$  = length of subterminal eupathidium on tarsus of the leg II;  $\sigma III$  = length of solenidion on genu of the leg III;  $\varphi III$  = length of solenidion on tibia of the leg III and MTa = length of mastisetae on tarsus of the leg III.

**Etymology.** The specific name *fernandae* is in the honor of the collector, Fernanda Martins Hatano, a professor of the Federal Rural University of Amazônia and a specialist in the ecology of rodent mites.

**Differential diagnosis.** *Parasecia fernandae* sp.n. is similar to *P. argentinensis* Goff et Gettinger, 1995, *P. soucouyanti* (Brennan et Yunker, 1966) and *P. valida* Brennan, 1969, in having mastiseta on tarsus of legs III and three σ on the genu of legs I. However, the new species differs from the above three in having a hypertrichous idiosoma (ca. 115 vs. 52–54, 48–56 and 42, respectively) and ε distal to ω (proximal in the other three species).

#### *Arisocerus hertigi* (Brennan et Jones, 1964)

This species was described based on specimens obtained from *Dasyprocta* sp. in Paraguay (Brennan and Jones 1964). In addition to this record, the species *A. hertigi* was reported in Brazil parasitizing *Didelphis albiventris* Lund, 1840 in the Federal District and *Cavia intermedia* Cherem, Olimpio and Ximenez, 1999 in Santa Catarina State (Goff et Gettinger 1989; Regolin et al. 2015). In this study, we reported the first record of *A. hertigi* parasitizing *N. squamipes* and *Oxymycterus* sp. and the first record to Rio de Janeiro State.

**Material examined.** 3 larvae (ACA-2613), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brazil, 15.III.2001, leg. F.M. Hatano. 1 larva (ACA-2620), same host, locality and collector, 15.V.2001. 1 larva (ACA-2622), same host, locality and collector, 16.III.2002. 1 larva (ACA-2627), ex. *Oxymycterus* sp., same locality and collector, 24.VIII.2002. 1 larva (ACA-2628), same host, locality and collector, 31.XI.2002. 1 larva (ACA- 2629) same host, locality and collector, 03.VI.2002. 1 larva (ACA-2630), same host, locality and collector, 31.XI.2002. 1 larva (ACA-2631), same host, locality and collector, 24.VIII.2002.

#### *Eutrombicula goeldii* (Oudemans 1910)

This species was found parasitizing several birds, reptiles and mammals in the following countries: Bolivia, Brazil, Colombia, Costa Rica, Dominica, Panama, Surinam, Trinidad, and Venezuela (Oudemans 1910; Boshell and Kerr 1942; Brennan and Jones 1960; Brennan and Yunker 1966; Brennan 1967; Brennan 1968; Arnold 1970; Brennan 1970a;

1970b; Brennan and Lukoschus 1971; Brennan and Reed 1974; Brennan and Bronswijk 1975). Brennan and Jones (1960) and Brennan and Reed (1974) recorded this chigger parasitizing a rodent misidentified as *N. squamipes* in Trinidad and Venezuela, respectively. Probably, the authors referred to the species *Nectomys palmipes* (Allen et Chapman, 1893), since, according Musser and Carleton (2005), this is the only species in the genus *Nectomys* that occurs in Trinidad and Venezuela. In Brazil, the only record of this species was from *Dasyprocta leporina* Linnaeus 1758 (=*Dasyprocta aguti* Linnaeus, 1766) with no detailed locality information. Here we are reporting the first record of *E. goeldii* parasitizing *N. squamipes* and the first record from Rio de Janeiro State.

**Material examined.** 1 larva (ACA-2626), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brasil, 14.II.2002, leg. F.M. Hatano.

#### *Microtrombicula brachytrichia* Brennan, 1971

The only record of this species is the type data: collected in Belém, Pará State, Brazil on *Proechimys guyannensis* (Geoffroy, 1803) (Brennan 1971). In this study, we are providing a new record of locality and host for this species.

**Material examined.** 1 larva (ACA-2633), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brazil, 30.VIII.2001, leg. F.M. Hatano. 2 larvae (ACA-2634), same data. 1 larva (ACA-2624b), same host, locality and collector, 16.II.2002.

#### *Parasecia manueli* (Brennan et Jones, 1960)

*Parasecia manueli* has been collected parasitizing some species of birds, reptiles, marsupials and rodents, in several countries of the Neotropical region: Colombia, Cuba, Panama, Peru, Surinam, Trinidad and Venezuela (Brennan and Jones 1960; 1961; Brennan and Yunker 1966; Brennan 1968; Brennan and Lukoschus 1971; Brennan and Bronswijk 1975; Brennan and Reed 1975; Daniel and Stekolnikov 2003). Brennan and Jones (1960) recorded this chigger parasitizing a rodent wrongly identified as *N. squamipes* in Trinidad, as well as *E. goeldii*. Probably the authors referred to the species *N. palmipes*. It is important to

note that *P. manueli* was recorded in Brazil for the first time in the present study, as well as, the first record of this mite parasitizing the rodent *N. squamipes*.

**Material examined.** 3 larvae (ACA-2616), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brazil, 17.III.2002, leg. F.M. Hatano. 1 larva (ACA-2618a), same data. 1 larva (ACA-2619), same host, locality and collector, 15.V.2001. 1 larva (ACA-2632), same host, locality and collector, II.2001. 2 larvae (ACA-2624a), same host, locality and collector, 16.III.2002.

#### ***Quadraseta brasiliensis* Goff et Gettinger, 1989**

The distribution of this species is restricted to the Brazilian territory, federal district and São Paulo and Paraná States (Goff and Gettinger 1989; Jacinavicius et al. 2018a). This species was found parasitizing several marsupials and rodents. However, it is the first time that it was found parasitizing *N. squamipes*, and it is the first record for Rio de Janeiro State.

**Material examined.** 1 larva (ACA-2614), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brazil, 14.V.2001, leg. F.M. Hatano. 2 larvae (ACA-2621), same host, locality and collector, 16.III.2002.

#### ***Serratacarus dietzi* Goff et Whitaker, 1984**

The only record of this species was the type data, Serra da Canastra National Park, Minas Gerais State, Brazil, parasitizing *Necromys lasiurus* (Lund, 1840) (Goff and Whitaker 1984). Here we are recording for the first time, these species parasitizing *N. squamipes* and *Cerradomys subflavus* (Wagner, 1842) in Rio de Janeiro State.

**Material examined.** 1 larva (ACA-2615), ex. *N. squamipes*, Parque Nacional Restinga de Jurubatiba, Macaé municipality, Rio de Janeiro State, Brasil, 02.VI.2002, leg. F.M. Hatano. 1 larva (ACA-2617), ex. *C. subflavus*, same locality and collector, 25.VIII.2002.

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**5. CHAPTER 4 – DESCRIPTION OF *Pseudoschoengastia petrolinensis* N. SP. (TROMBIDIFORMES: TROMBICULIDAE), AND NEW RECORDS OF CHIGGERS FROM NORTHEASTERN BRAZIL**

**5.1 INTRODUCTION.**

The mites belonging to the families Trombiculidae and Leeuwenhoekiidae are popularly known as “chiggers”, and during the larval stage they are parasites of vertebrates (Ewing 1944). According to Zhang et al. (2011) approximately 3,700 species have been described worldwide, of these 66 species have been recorded from Brazil (Bassini-Silva et al. 2018a; Jacinavicius et al. 2018a, b). Only one species of chiggers, *Apolonia tigipioensis* Torres et Braga, 1938 (Trombidiformes: Leeuwenhoekiidae), has been recorded from the state of Pernambuco which was collected parasitizing birds and humans (Torres and Braga 1938; Carneiro 1949, 1952; Ornelas-Almeida et al. 2007; Bassini-Silva et al. 2018b).

The genus *Pseudoschoengastia* Lipovsky, 1951 (Trombidiformes: Trombiculidae) is widely distributed in Neotropical and Nearctic regions with 29 described species from Colombia, Costa Rica, French Guiana, Guatemala, Mexico, Panama and USA (Hoffmann 1948, 1951, 1960; Lipovsky 1951; Brennan 1952, 1960, 1965, 1968; Brennan and Jones 1959; Fauran 1960; Brennan and Yunker 1966; Geest and Loomis 1968; Loomis 1976; Goff 1982; Suzuki and Mamiya 1982). A new species, *Pseudoschoengastia petrolinensis* n. sp. was discovered parasitizing a white-eared opossum, *Didelphis albiventris* Lund (Didelphimorphia: Didelphidae), in Carneiros, Petrolina municipality, Pernambuco state, Brazil. In addition, new records for the following species are provided: *Eutrombicula batatas* (Linnaeus, 1758), *Eutrombicula spipi* Brennan and Reed, 1974 and *Quadraseta falconensis* Goff and Brennan, 1977.

**5.2 MATERIALS AND METHODS**

Chiggers were collected in the ears of nine *D. albiventris* from Carneiros, a rural zone in the municipality of Petrolina, Pernambuco State, Brazil. The collections occurred on the banks of the São Francisco River, in a semi-arid region of the Caatinga biome, in May 2015. The collections were carried out on license nº SisBio - 45764-1, coordinated by Mauricio C. Horta.

Marsupials were captured using Tomahawk (45 x 16 x 16 cm) traps, which were baited with grits, sardines, bagels, bananas and pineapples and placed in shaded areas near trees. Physical restraint was applied with the help of gloves, and then weighing and subsequent chemical restraint were applied using 15-30 mg/kg of ketamine hydrochloride (Cubas et al. 2014). Additionally, ectoparasites were manually collected from the wild animals and stored in 1.5 mL microtubes containing absolute ethyl alcohol (C<sub>2</sub>H<sub>5</sub>OH) at –20°C prior to the laboratory analysis. After the collection of biological materials, the animals were marked by cutting the hair from the sacral region and were liberated from the same point of capture after full anesthesia recovery.

All mites, previously stored in absolute alcohol, were slide-mounted in Hoyer's medium (Walter and Krantz 2009) and identified at the genus level using a key published by Brennan and Goff (1977). Subsequently, specimens were identified to the species level by comparing them to the original description of all species of the genus and when is possible, with type specimens that were deposited at the USNM, which are housed at the Systematic Entomology Laboratory (USDA-ARS, BARC).

Illustrations of the morphological characters and the study of types were done using a Leica microscope DM4000B, with a camera lucida. Extended focal range images were compiled with the Leica Application Suite version 2.5.0. The SEM micrographs were obtained with a Digital Scanning Microscope FEI, Quanta 250, located at the Laboratório de Biologia Celular, Instituto Butantan. The images were prepared with Adobe Photoshop v. 13.0 and Inkscape V.2. We also used the nomenclature proposed by Grandjean (1939) with adaptations by Kethley (1990) for the dorsal opisthosomal setae and setae on the prodorsal sclerite and Grandjean (1935, 1947) for the specialized setae on the legs and palp. All the specimens were deposited in the Acari Collection of the Instituto Butantan, São Paulo, Brazil (IBSP).

### 5.3 RESULTS

The following four species of chiggers were collected in the study: *Eutrombicula batatas* (Linnaeus, 1758); *Eutrombicula spipi* Brennan and Reed, 1974; *Quadraseta falconensis* Goff and Brennan, 1977 and a new species of the genus *Pseudoschoengastia* described herein. We also observed one host was co-parasitized with *Pseudoschoengastia petrolinensis* n. sp., *E. batatas* and *Q. falconensis*; and another host had *Pseudoschoengastia*

*petrolinensis* n. sp., and *Q. falconensis*. This is the first record of *Pseudoschoengastia* from Brazil, increasing the number of the species to the Neotropical region and enlarging the distribution area. The species *E. spipi* and *Q. falconensis* are also new records to Brazil. The marsupial *D. albiventris* is a new host record to *E. batatas*, *E. spipi*, *Q. falconensis*, and the state of Pernambuco is a new locality for all these species.

### Family Trombiculidae Ewing, 1944

#### Genus *Pseudoschoengastia* Lipovsky, 1951

Type species: *Pseudoschoengastia hungerfordi* Lipovsky, 1951: 95, by original designation.

**Diagnosis.** Larva. Five branched setae on palptarsus, without eupathidium ( $\zeta$ ); odontus trifurcated; cheliceral blade with tricuspid cap; prodorsal sclerite with 1 pair of expanded internal scapular trichobothridial setae (*si*), *se* setae off the prodorsal sclerite; femora of legs I divided into a basifemur and telofemur, leg II and III divided into a basifemur and telofemur or fused into a femur; empodium present and onychotriches absent; Leg I and II with subterminal eupathidia; tarsus of the leg I with a dorsal eupathidium ( $\zeta$ ), a companion seta (*z*) and a subterminal eupathidium ( $\zeta$ ); mastiseta on tarsus of the leg III absent.

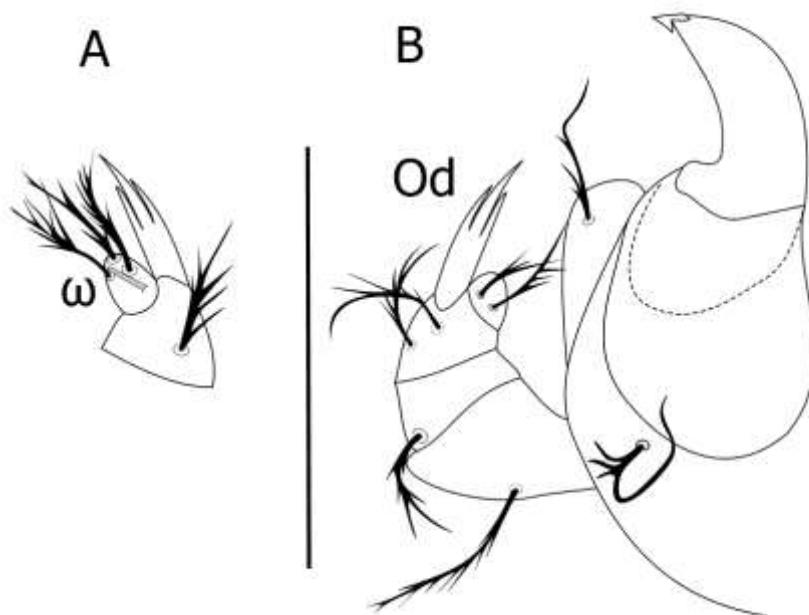
#### *Pseudoschoengastia petrolinensis* Jacinavicius, Bassini-Silva and Barros-Battesti n. sp.

#### Figs. 1-5; Table 1, 2

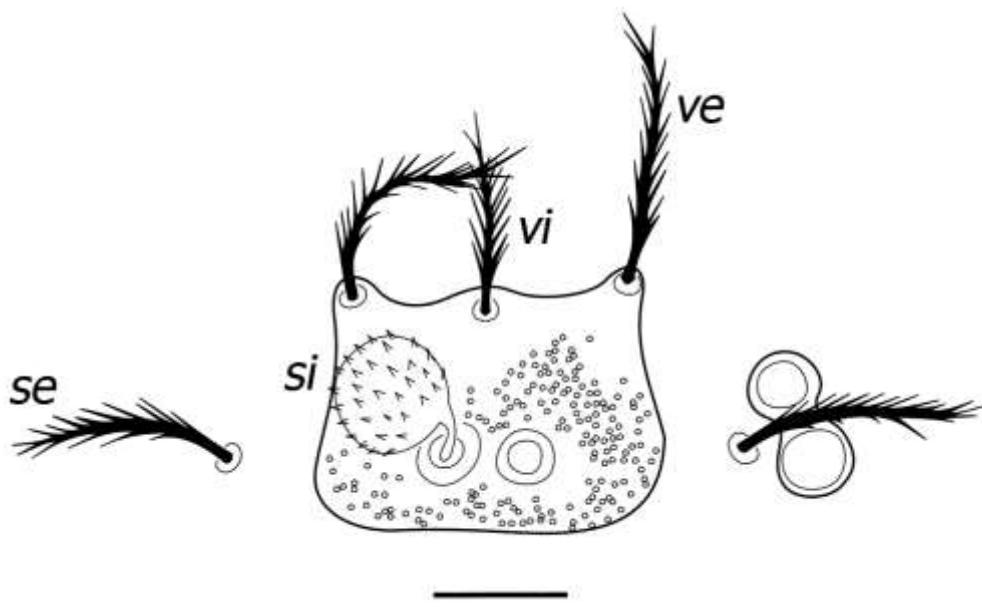
**Diagnosis.** Palptibia with the dorsal, lateral and ventral branched setae, adoral setae branched, idiosoma with ca. 90 setae; genu of the legs I each with three  $\sigma$ , tarsus of the legs I and II with a  $\epsilon$  proximal to  $\omega$ .

**Description (holotype).** Larva. *Gnathosoma* - palp setal formula B/B/BBB/5B $\omega$ ; odontus trifurcate, supracoxal setae branched, cheliceral blade with tricuspid cap; adoral setae (galeal setae) branched (Figs. 1A-B). *Idiosoma* - eyes 2/2 set in an ocular plate, posterior eye larger than the anterior eye; prodorsal sclerite with 1 pair of globose internal scapular trichobothridial setae (*si*), a single *vi* (= AM) seta, 1 pair of *ve* (= AL) seta and one pair of *se* (= PL) seta off the prodorsal sclerite; *si* > *vi* > *se* > *ve*; the anterior margin of the prodorsal sclerite biconcave, and the lateral margins slightly concave (Fig. 2). Idiosoma with 97 setae, dorsal opisthosoma with extra setae in the C and H rows, 5 pairs of setae in the C row, and the  $c_5$  pair of seta in an anterior position (= humeral setae), D row with 3 pairs of setae, E row with 3 pairs of setae, F row with 3 pairs of setae, H row with 5 pairs of setae, totaling 38

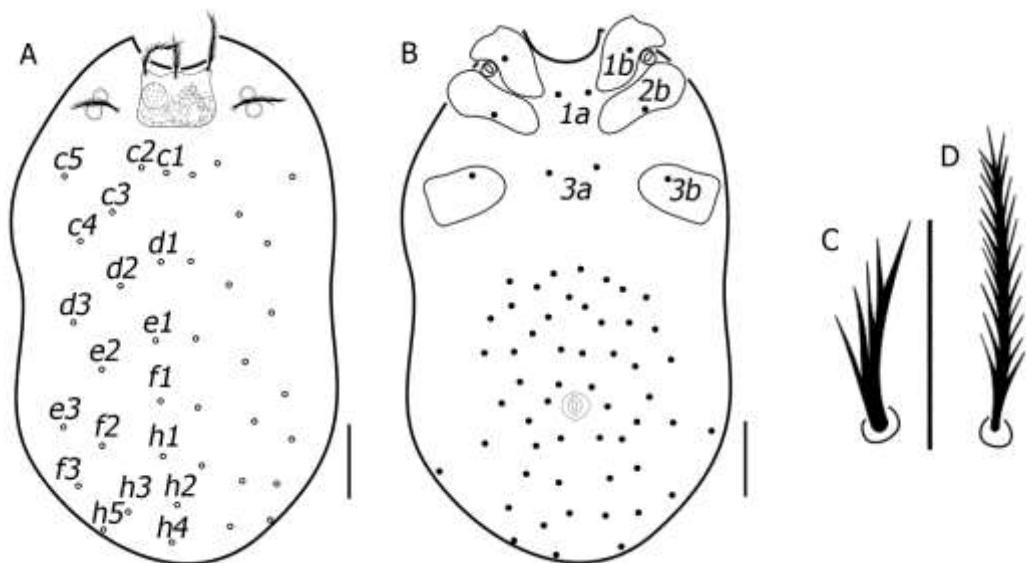
dorsal opisthosomal setae, 2 pairs of sternal setae (1a, 3a), and 30 setae located anterior of the anus and 23 setae located posterior of the anus, totaling 53 ventral setae (Figs. 3A-D, 5A and C). *Legs* - femur legs I-III each divided into a basifemur and telofemur, each leg terminating with a pair of claws and a claw-like empodium, without onychotriches, coxal fields not striate. *Leg I* - coxal field with 1 branched seta 1b (1B); trochanter 1B; basifemur 1B; telofemur 5B; genu 4B, σ 3, with κ; tibia 8B, φ 2, with κ; tarsus 21B, with ω, ε, dorsal eupathidium ( $\zeta$ ) with a companion seta (z) and terminating with a subterminal eupathidium ( $\zeta$ ), famulus ( $\varepsilon$ ) proximal to  $\omega$  (Fig. 4A, 5D). *Leg II* - coxal field seta 2b (1B); trochanter 1B; basifemur 2B; telofemur 4B; genu 3B, σ; tibia 6B, φ 2; tarsus 17B, with  $\omega$ , ε, and a subterminal eupathidium ( $\zeta$ ), base of ε proximal to  $\omega$  (Fig. 4B, 5B). *Leg III* - coxal field seta 3b (1B), trochanter 1B; basifemur 2B; telofemur 3B; genu 3B, σ; tibia 6B, φ; tarsus 15B (Fig. 4C, 5C).



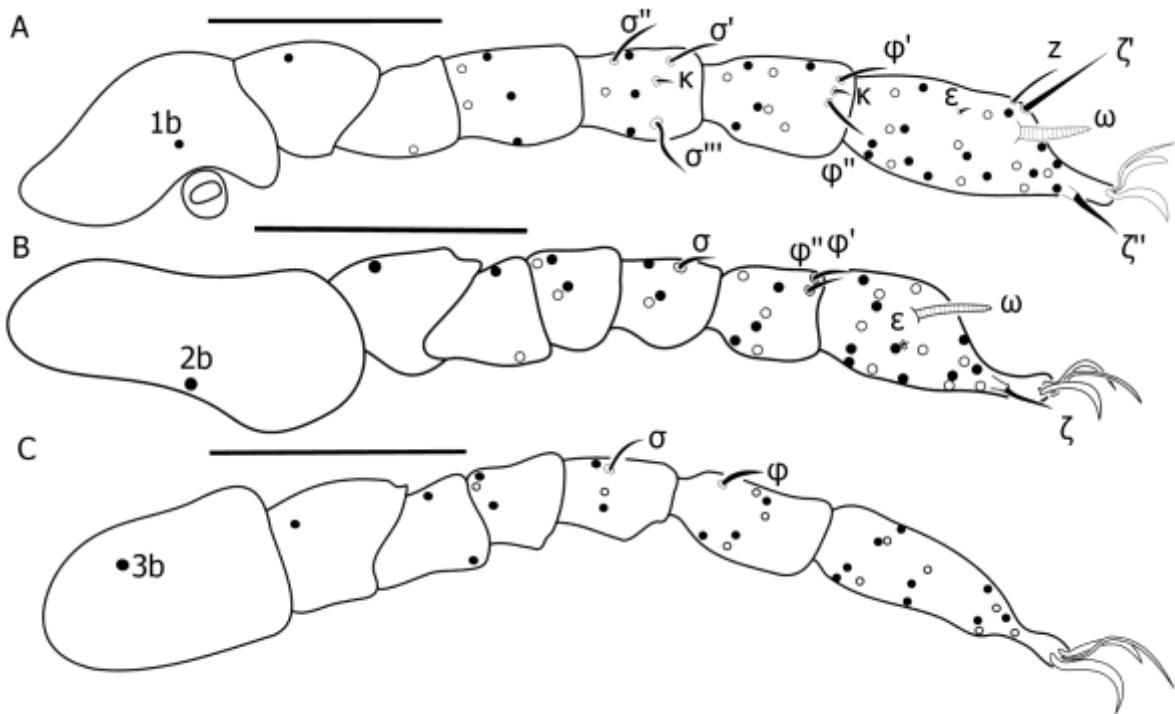
**Figure 1.** Morphological details of *Pseudoschoengastia petrolinensis* n. sp. larva. A. ventral view of palptarsus; B. dorsal view of gnathosoma; Symbols:  $\omega$  = solenidion of palptarsus; Od = odontus; Scales: A-B 50 $\mu$ m.



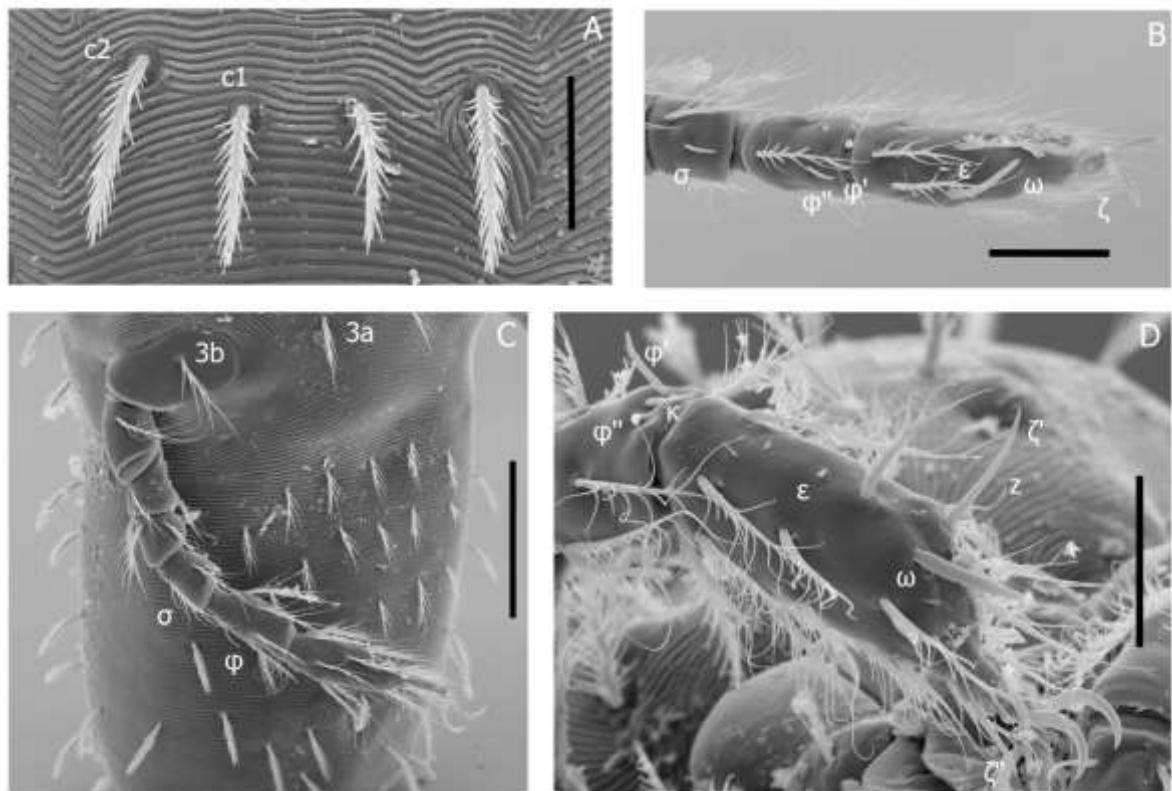
**Figure 2.** Morphological details of *Pseudoschoengastia petrolinensis* n. sp. larva. A. prodorsal sclerite; Symbols: *ve* = external vertical setae; *se* = external scapular setae; *vi* = internal vertical setae; *si* = internal scapular setae or trichobothria; Scales: 20 $\mu$ m.



**Figure 3.** Morphological details of *Pseudoschoengastia petrolinensis* n. sp. larva. A. dorsal view of idiosoma; B. ventral view of idiosoma; C. dorsal opisthosomal setae; D. ventral opisthosomal setae. Black spots = ventral setae of the idiosoma; white spots = dorsal setae of the idiosoma. Symbols: *c<sub>1</sub>-c<sub>5</sub>* = setae of the C row; *d<sub>1</sub>-d<sub>3</sub>* = setae of the D row; *e<sub>1</sub>-e<sub>3</sub>* = setae of the E row; *f<sub>1</sub>-f<sub>3</sub>* = setae of the F row; *h<sub>1</sub>-h<sub>5</sub>* = setae of the H row; *1a* = anterior sternal setae; *3a* = posterior sternal setae; *1b* = seta of the coxal field of the leg I; *2b* = seta of the coxal field of the leg II; *3b* = seta of the coxal field of the leg III; Scales: A and B 50 $\mu$ m; C and D 20 $\mu$ m.



**Figure 4.** Morphological details of *Pseudoschoengastia petrolinensis* n. sp. larva. A. Leg I; B. Leg II; C. Leg III; Black spots = ventral setae of the legs; white spots = dorsal setae of the legs. Symbols:  $\zeta'$  = dorsal eupathid of the tarsus of the leg I;  $\zeta''$ ,  $\zeta$  = subterminal eupathid of the tarsus of the leg I and II, respectively;  $\omega$  = solenidion of the tarsus on the leg I and II;  $\sigma$ ,  $\sigma'$ ,  $\sigma''$  and  $\sigma'''$  = solenidia on the genu of the legs I, II and III;  $\kappa$  = microsetae on genu and tibia of the leg I;  $\varphi'$ ,  $\varphi''$ ,  $\varphi$  = solenidia on the tibia of the legs I, II and III;  $\varepsilon$  = famulus on the tarsus of the legs I and II;  $z$  = companion seta on tarsus of the leg I;  $2b$  = seta of the coxal field of the leg II;  $3b$  = seta of the coxal field of the leg III; Scales: A-C 50 $\mu$ m.



**Figure 5.** Scanning electron micrographs of *Pseudoschoengastia petrolinensis* n. sp. larva. A. dorsal opisthosomal setae. B. tarsus, tibia and genu of the leg II. C. ventrolateral view of the idiosoma. D. tarsus of the leg I. Symbols:  $c_1$  and  $c_2$  = setae of the C row;  $\zeta$  = eupathidium of palptarsus;  $\zeta'$  = dorsal eupathidium of the tarsus of the leg I;  $\zeta''$ ,  $\zeta$ , = subterminal eupathidium of the tarsus of the leg I and II, respectively;  $\omega$  = solenidion of palptarsus and tarsus of the leg I and II;  $\sigma$  = solenidion on the genu of the legs II and III;  $\kappa$  = microsetae on tibia of the leg I;  $\phi'$   $\phi''$  = solenidia on the tibia of the legs I, II and III;  $\varepsilon$  = famulus on the tarsus of the legs I and II;  $z$  = companion seta on tarsus of the leg I;  $3a$  = posterior sternal setae;  $3b$  = seta of the coxal field of the leg III; Scales: A, B and D 20 $\mu$ m; C 50 $\mu$ m.

**Table 1.** Standard measurements of *Pseudoschoengastia petrolinensis* sp. n. (n = 26)

	AW	PW	SB	ASB	PSB	SD	P-PL	AP	vi	ve	se	si	c <sub>5</sub>	1a	3a	DMIN	DMAX	VMIN	VMAX
<b>Holotype</b>	41	76	11	25	11	36	11	30	27	41	33	22	35	24	23	22	27	11	22
Minimum	41	73	10	25	11	36	11	27	27	40	33	22	35	23	23	22	27	11	22
Maximum	44	76	11	28	17	45	18	30	34	45	35	24	36	24	24	23	30	15	22
Mean	42	74	11	26	14	40	16	29	31	42	34	23	35	24	23	22	29	13	22

Legend: AW = distance between the bases of the *ve* setae; PW = distance between the bases of the *se* setae; SB = distance between bothridial bases; ASB = distance from bothridial bases to extreme anterior margin of the prodorsal sclerite; PSB = distance from bothridial bases to extreme posterior margin of the prodorsal sclerite; SD = ASB + PSB; P-PL = distance from posterolateral to extreme posterior margin; AP = distance between the bases of *ve* and *se*; vi = anteromedian seta; *ve* = anterolateral setae; *se* = posterolateral setae; *si* = trichobothria; *c<sub>5</sub>* = humeral setae; 1a = anterior sternal setae; 3a = posterior sternal setae; DMIN = minimum length of dorsal opisthosomal setae; DMAX = maximum length of dorsal opisthosomal setae; VMIN = minimum length of ventral idiosomal setae and VMAX = maximum length of ventral idiosomal setae.

**Table 2.** Standard measurements of the specialized setae of *Pseudoschoengastia petrolinensis* n. sp. (n=26)

	I	II	III	Ip	TaIII	TaW	$\sigma$ I	$\kappa$ I	$\phi$ I	$\kappa$ I	$\omega$ I	$\varepsilon$ I	$\zeta'$ I	$\zeta''$ I	z	$\sigma$ II	$\phi$ II	$\omega$ II	$\varepsilon$ II	$\zeta$ II	$\sigma$ III	$\phi$ III
<b>Holotype</b>	223	167	203	593	54	14	13	4	12	3	14	2	10	18	11	8	8	14	3	10	10	9
Minimum	212	167	203	589	51	14	11	4	11	3	12	2	10	18	10	7	7	14	3	9	8	9
Maximum	223	180	210	604	54	16	13	4	12	3	14	2	10	20	11	8	8	15	3	10	10	10
Mean	216	174	205	179	53	15	12	4	12	3	13	2	10	19	11	8	7	15	3	9	9	9

Legend: I = length of leg I; II = length of leg II; III = length of leg III; Ip = sum of leg lengths (coxal field to tarsus); TaIII = length of tarsus of the leg III; TaW = width of tarsus of the leg III;  $\sigma$  I = length of an average of the three solenidia on genu of the leg I;  $\kappa$  I = length of microseta on genu of the leg I;  $\phi$  I = length of an average of both solenidia on tibia of the leg I;  $\kappa$  I = length of microseta on tibia of the leg I;  $\omega$  I = length of solenidion on tarsus of the leg I;  $\varepsilon$  I = length of famulus on tarsus of the leg I;  $\zeta'$  I = length of dorsal eupathidium on tarsus of the leg I;  $\zeta''$  I = length of subterminal eupathidium on tarsus of the leg I; z = length of companion seta on tarsus of the leg I;  $\sigma$  II = length of solenidion on genu of the leg II;  $\phi$  II = length of an average of both solenidia on tibia of the leg II;  $\omega$  II = length of solenidion on tarsus of the leg II;  $\varepsilon$  II = length of famulus on tarsus of the leg II;  $\zeta$  II = length of subterminal eupathidium on tarsus of the leg II;  $\sigma$  III = length of solenidion on genu of the leg III and  $\phi$  III = length of solenidion on tarsus of the leg III.

**Type data.** Holotype larva (IBSP 13045), Carneiros, Petrolina municipality, Pernambuco state, Brazil ( $40^{\circ} 26' 16.7''$  W,  $09^{\circ} 21' 15.3''$  S, 360m), 01-V-2015, ex. *Didelphis albiventris*, M. C Horta coll. Paratypes, same data: 4 larvae (IBSP 13045). Holotype and paratypes were deposited in the IBSP collection.

**Additional material examined.** 2 larvae (IBSP 13038), Carneiros, Petrolina municipality, Pernambuco state, Brazil ( $40^{\circ} 26' 7.9''$  W,  $09^{\circ} 21' 3.3''$  S, 371m), 01-V-2015, ex. *Didelphis albiventris*, M. C Horta coll.; 2 larvae (13039A), same data; 1 larva (IBSP 13040) ( $40^{\circ} 26' 11.5''$  W,  $09^{\circ} 21' 5.8''$  S, 366m), same data; 4 larvae (IBSP 13041), same data; 2 larvae (IBSP 13042A) ( $40^{\circ} 26' 12.2''$  W,  $09^{\circ} 21' 8.9''$  S, 365m), same data; 4 larvae (IBSP 13043), same data; 6 larvae (IBSP 13044) ( $40^{\circ} 26' 10.3''$  W,  $09^{\circ} 21' 6.9''$  S, 364m), same data.

**Etymology.** The specific epithet *petrolinensis* refers to the type locality of the species, Petrolina municipality.

**Differential diagnosis.** The species *Pseudoschoengastia petrolinensis* n. sp. is most similar to *P. bisetosa* Loomis, 1976 and *P. mexicoensis* Susuki et Kamiya, 1982 in having adoral setae and all of setae of the palptibia branched, but differs because both species have only two  $\sigma$  on the genu of the leg I and  $\epsilon$  distal to  $\omega$  on the tarsus of the leg I, while the new species has three  $\sigma$  on the genu of the leg I and  $\epsilon$  is proximal to  $\omega$  on the tarsus of the leg I. Besides that *P. bisetosa* has two setae on coxa of the legs II and III (vs. one seta) and *P. mexicoensis* has a clavate trichobothria (vs. trichobothria globose). In addition, the length of  $ve$  (40-45) of *P. petrolinensis* n. sp. is greater than *P. bisetosa* and *P. mexicoensis* (20-32 and 20, respectively), besides that the new species has PW greater (73-76) and smaller  $se$  (33-35) than *P. mexicoensis* (58-63 and 40-43, respectively).

***Eutrombicula batatas* (Linnaeus, 1758)**

*Acarus batatas* Linnaeus, 1758: 617;

*Trombicula brasiliensis* Ewing, 1925: 92;

*Trombicula flui* van Thiel, 1930:347;

*Trombicula pastora* Boshell and Kerr, 1942:119;

*Acariscus hominis* Ewing, 1943:63;

*Trombicula (Eutrombicula) batatas*: Jenkins 1949:298; Brennan 1953: 294; Brennan and Dalmat 1960: 188;

*Eutrombicula batatas*: Brennan and Jones 1960: 508; Brennan and Reed 1974: 709; Brennan and Reed 1975: 51; Hoffmann, 1990: 55; Faccini et al. 2017:105.

This species is widespread in the Neotropical region, occurring in Argentina, Bolivia, Brazil, Colombia, Costa Rica, Honduras, Curaçao, Guatemala, French Guyana, Jamaica, Mexico, Panama, Paraguay, Puerto Rico, Surinam, Trinidad, USA and Venezuela, where it parasitizes

birds, reptiles and mammals, including humans (Hoffmann 1990; Estébanes-González and Cervantes 2005; Faccini et al. 2017; Bassini-Silva et al 2018a).

**Material examined.** 1 larva (IBSP 13039B), Carneiros, Petrolina municipality, Pernambuco state, Brazil ( $40^{\circ} 26' 13.4''$  W,  $09^{\circ} 21' 2''$  S, 372m), 01-V-2015, ex. *Didelphis albiventris*, M. C Horta coll.

**Type specimens examined.** *Trombicula brasiliensis* Ewing, 1925 (junior synonym of *E. batatas*) – Holotype (USNM 946), Manaus municipality, Brazil, 25-VII-1924.

***Eutrombicula spipi*** Brennan and Reed, 1974

*Eutrombicula spipi* Brennan and Reed, 1974: 705; Brennan and Reed 1975: 52.

This species was originally in Venezuela, from the rodent *Makalata didelphoides* (Desmarest, 1817) (Syn.: *Echimys armatus* Geoffroy, 1838) (Rodentia: Echimyidae) and two marsupials *Lutreolina crassicaudata* (Desmarest, 1804) (Didelphimorphia: Didelphidae) and *Marmosops fuscatus* Thomas, 1896 (Didelphimorphia: Didelphidae) (Brennan and Reed 1974).

**Material examined.** 5 larvae (IBSP 13046B), Carneiros, Petrolina municipality, Pernambuco state, Brazil ( $40^{\circ} 26' 11.5''$  W,  $09^{\circ} 21' 5.8''$  S, 366m), 01-V-2015, ex. *Didelphis albiventris*, M. C Horta coll.

**Type species examined.** Holotype and 9 paratypes (RML 52734), Amazonas, Venezuela, 6-III-1967, ex. *Echimys armatus* Geoffroy, 1838, housed at the NMNH.

***Quadraseta falconensis*** Goff and Brennan, 1977

*Quadraseta falconensis* Goff and Brennan, 1977: 505

This species has only been recovered from *Sylvilagus floridanus* (Allen, 1890) (Lagomorpha: Leporidae) in Venezuela (Goff and Brennan 1977).

**Material examined.** 1 larva (IBSP 13039C), Carneiros, Petrolina municipality, Pernambuco state, Brazil ( $40^{\circ} 26' 13.4''$  W,  $09^{\circ} 21' 2''$  S, 372m), 01-V-2015, ex. *Didelphis albiventris*, M. C Horta coll. 1 larva (IBSP 13042B), same data.

**Type species examined.** Holotype and 7 paratypes (RML 53360), Coro municipality, Falcon state, Venezuela, 17- VII-1968, *Sylvilagus floridanus*.

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**6. CHAPTER 5 – SYNONYMY OF THE GENUS *Arisocerus* BRENNAN, 1970  
WITH THE GENUS *Herpetacarus* VERCAMMEN-GRANDJEAN, 1960  
(TROMBIDIFORMES: TROMBICULIDAE)**

**6.1 INTRODUCTION**

The genus *Arisocerus* Brennan, 1970 is one of the chigger genera that have been reported on the Neotropical region, including two species, the type species of the genus, *Arisocerus amapensis* Brennan, 1970 and the species *Arisocerus hertigi* (Brennan & Jones, 1964). The specific epithet of the first species refers to the type locality, Amapá state, Brazil and other was in honor to the collector, Dr Marshall Hertig, entomologist of the Gorgas Memorial Laboratory (The Gorgas Memorial Institute for Health Studies), Panama (Brennan 1970).

Here we present the genus *Arisocerus* as a junior synonym of the genus *Herpetacarus* (*Herpetacarus*), including a redescription of the species *Herpetacarus* (*Herpetacarus*) *hertigi* **com. nov.** and *Herpetacarus* (*Herpetacarus*) *amapensis* **com. nov.**. In additional, new records of locality from Brazil and host for *H. (H.) hertigi* **com. nov.** has been provided.

**6.2 MATERIAL AND METHODS**

Slide-mounted larvae of chiggers housed at the collection at the NMNH, which are housed at the Systematic Entomology Laboratory (BARC-USDA-ARS), Washington, EUA, were examined. In addition, chiggers housed at the Acari Collection of the Instituto Butantan (IBSP) was part slide-mounted with Hoyer's medium according to Walter and Krantz (2009), part was also prepared for Scanning Electron Microscopy (SEM) according to Walter and Krantz (2009) with a Digital Scanning Microscope FEI, Quanta 250, at the Laboratory of Cell Biology, Butantan Institute, for identification.

The morphological illustrations of all species were made using Leica DFC 500 digital camera coupled to an optical microscope Leica DM4000B. Extended focal range images were composed with Leica Application Suite version 2.5.0. The distribution map was prepared using the DIVAGIS program. The images were prepared with Adobe Photoshop v. 13.0, and Inkscape V.2.

For descriptions the specimens, was used the terminology of Goff *et al.* (1982) with adaptations (Stekolnikov 2008, Stekolnikov & Daniel 2012) and also adopted the

nomenclature proposed by Kethley (1990), Wohltmann *et al.* (2006); Wohltmann *et al.* (2007), and Bassini-Silva *et al.* (2016). If necessary, the current host name was listed with the original host name in parenthesis. The host species were updated based on the IUCN (2019).

### 6.3 SYSTEMATICS

#### **Family Trombiculidae Ewing, 1944**

##### **Genus *Herpetacarus* (*Herpetacarus*) Vercammen-Grandjean, 1960**

*Herpetacarus* (*Lukoschuskaaia*) Kolebinova and Vercammen-Grandjean 1980: 56, Stekolnikov 2018: 65.

*Ornithacarus* Vercammen-Grandjean 1960: 469; Vercammen-Grandjean 1966: 631.

*Herpetacarus* (*Herpetacarus*) Vercammen-Grandjean 1960: 469; Fernandes and Kulkarni 2003: 366; Chung *et al.* 2015: 1249; Stekolnikov 2018:65.

*Arisocerus* Brennan 1970: 32; Brennan and Reed 1975: 49; Brennan and Goff 1977: 556; **syn. nov.**

**Type-species:** *Ascoshochengastia causicola* Jadin and Vercammen-Grandjean, 1952

**Diagnosis.** Larva. Palptarsus with seven branched setae (plus  $\omega$ ,  $\zeta$ ), odontus trifurcate;  $se$  on the prodorsal sclerite;  $si$  expanded; eyes 2/2 set in an ocular plate; femur of the legs I-III each divided into a basifemur and telofemur ; each leg terminated with a pair of claws and a clawlike empodium, without onychotriches; each genu of the legs I with two or three  $\sigma$ ; each genu of the legs II and III with one  $\sigma$ ; tibia from the leg III with one  $\varphi$ ; each tarsus of the leg I with a dorsal eupathid, a companion seta and terminating with a subterminal eupathid; presence or absence of mastisetae on tarsus of the leg III.

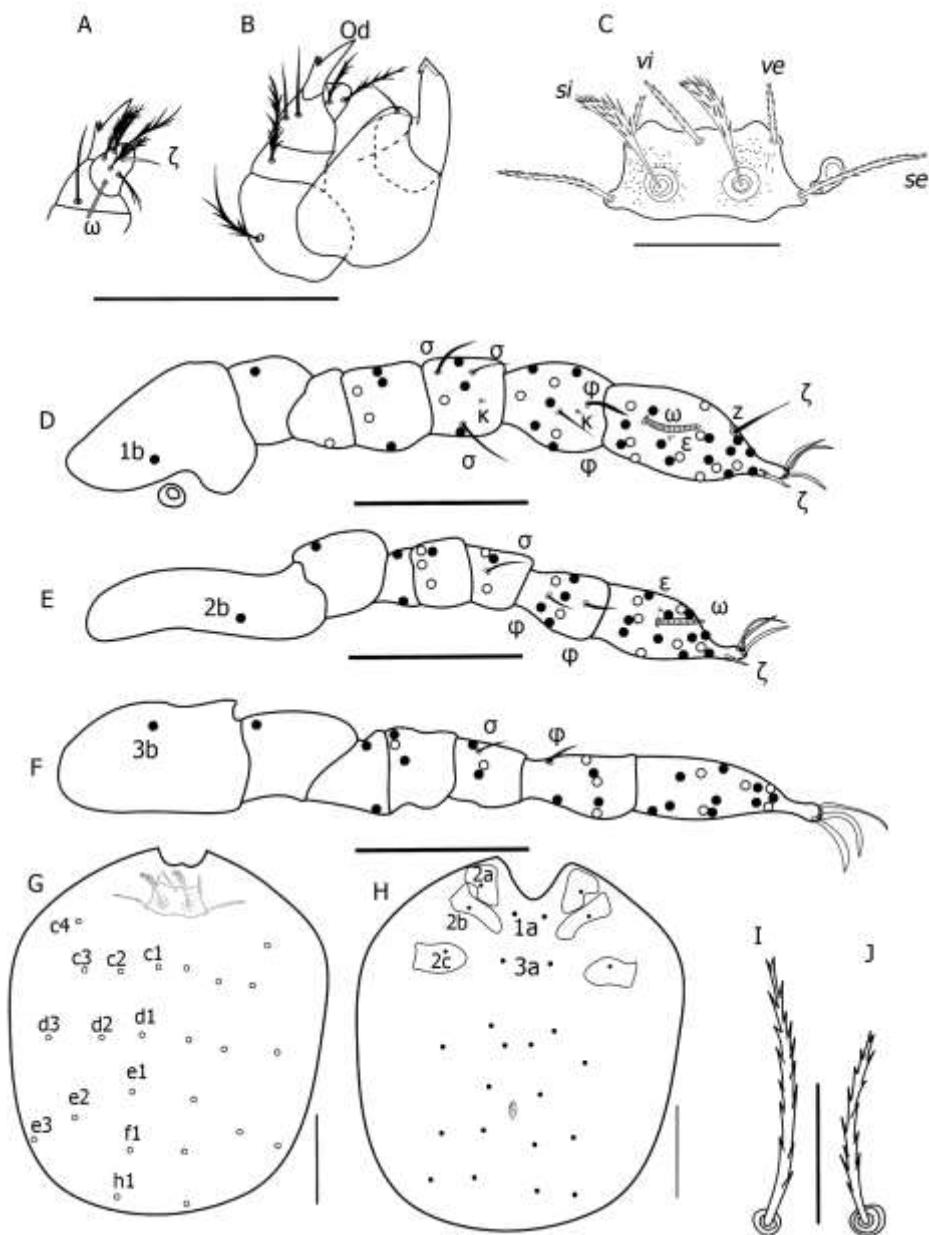
Postlarval instars: Unknown.

##### ***Herpetacarus* (*Herpetacarus*) *amapensis* (Brennan, 1970) com. nov. (Fig. 1)**

*Arisocerus amapensis* Brennan 1970: 32; Brennan and Reed 1975: 49; Brennan and Bronswijk 1975: 243; Jacinavicius *et al.* 2018a: 5.

*Herpetacarus* (*Herpetacarus*) *amapensis* com. nov.

**Diagnosis.** Larva. Palptibia with the dorsal, lateral and ventral nude setae; adoral setae nude; idiosoma with 44 setae; tarsus of the legs I with  $\epsilon$  distal to  $\omega$ , tarsus of the leg II with  $\epsilon$  in the same position to  $\omega$ , tarsus of the legs III without mastisetae.



**Figure 1.** Morphological details of *Herpetacarus (Herpetacarus) amapensis* com. nov. larva. A. ventral view of palptarsus; B. dorsal view of gnathosoma; C. prodorsal sclerite; D. Leg I; E. Leg II; F. Leg III; G. dorsal view of idiosoma; H ventral view of idiosoma; I. dorsal opisthosomal setae; J. ventral opisthosomal setae. Symbols: ve = external vertical setae; se = external scapular setae; vi = internal vertical setae; si = internal scapular setae or trichobothria; ω = solenidion of palptarsus and of the tarsus on the leg I and II; Od = odontus; ζ = dorsal eupathid of the tarsus of the leg I and subterminal eupathid of the tarsus of the leg I and II and of the palptarsus; σ = solenidia on the genu of the legs I, II and III; κ = microsetae on genu and tibia of the leg I; φ = solenidia on the tibia of the legs I, II and III; ε = famulus on the tarsus of the legs I and II; z = companion seta on tarsus of the leg I; 2b = seta of the coxal field of the leg II; 3b = seta of the coxal field of the leg III; c<sub>1</sub>-c<sub>4</sub> = setae of the C row; d<sub>1</sub>-d<sub>3</sub> = setae of the D row; e<sub>1</sub>-e<sub>3</sub> = setae of the E row; f<sub>1</sub> = setae of the F row; h<sub>1</sub> = setae of the H row; 1a = anterior sternal setae; 3a = posterior sternal setae; Black spots = ventral setae of the idiosoma; white spots = dorsal setae of the idiosoma. Scales: A-H 50µm; I and J 20µm.

**Redescription (Holotype and Paratypes).** Larvae. *Gnathosoma* - palp setal formula B/B>NNN/7B $\zeta\omega$ ; odontus trifurcate, supracoxal setae branched, cheliceral blade with tricuspid cap; adoral setae nude (Figs. 1A-B). *Idiosoma* - eyes 2/2 set in an ocular plate, anterior eye larger than the posterior eye; prodorsal sclerite with 1 pair of expanded internal scapular trichobothridial setae (*si*) with long and slender setules, a single *vi* (= AM) seta, 1 pair of *ve* (= AL) seta and one pair of *se* (= PL) seta on the prodorsal sclerite; *se* > *si* > *vi* > *ve*; the anterior and posterior margin of the prodorsal sclerite undulating, and the lateral margins slightly concave (Fig. 1C). Idiosoma with 44 setae, 4 pairs of setae in the C row, and the c<sub>4</sub> pair of seta in an anterior position (= humeral setae), D row with 3 pairs of setae, E row with 3 pairs of setae, F row with 1 pair of setae, H row with 1 pair of setae, totalizing 24 dorsal opisthosomal setae, 2 pairs of sternal setae (1a, 3a), and 8 setae located anterior of the anus and 8 setae located posterior of the anus, totalizing 16 ventral setae (Figs. 1G-J). Legs - femur legs I-III each divided into a basifemur and telofemur, each leg terminating with a pair of claws and a claw like empodium, without onychotriches, coxal fields not striate. *Leg I* - coxal field with 1 branched seta *1b* (1B); trochanter 1B; basifemur 1B; telofemur 5B; genu 4B, 3 σ, with κ; tibia 8B, 2 φ, with κ; tarsus 21B, with ω, ε, dorsal eupathid (ζ) with a companion seta (z) and terminating with a subterminal eupathid (ζ), famulus (ε) distal to ω (Fig. 1D). *Leg II* - coxal field seta *2b* (1B); trochanter 1B; basifemur 2B; telofemur 4B; genu 3B, σ; tibia 6B, 2 φ; tarsus 16B, with ω, ε, and a subterminal eupathid (ζ), base of ε in the same position to ω (Fig. 1E). *Leg III* - coxal field seta *3b* (1B), trochanter 1B; basifemur 2B; telofemur 3B; genu 3B, σ; tibia 6B, φ; tarsus 14B (Fig. 1F).

**Type and material examined.** PARATYPES: 5 larvae (RML 49368), Serra do Navio, Amapa, 15-III-1968, *Euryoryzomys macconnelli*; 6 larvae (RML 49382), same locality and host, 1-II-1968, 2 larvae (RML 49331), same data. Material: BRAZIL – 7 larvae (RML 49346), Serra do Navio, Amapa, 14-II-1968, *Euryoryzomys macconnelli*; 11 larvae (RML 49361), same locality, 9-III-1968, *Hylaeamys megacephalus* (= *Oryzomys capito*); 1 larva (RML 49092), same locality and host, 1-IV-1968; 1 larva (RML 49339), same locality and host, 9-II-1968; 1 larva (RML 49334), same locality and host, 5-II-1968; 2 larvae (RML 49345), same locality and host, 16-II-1968; 3 larvae (RML 49363), same locality and host, 11-III-1968; 2 larvae (RML 51726), Catu Forest (IPEAN), Belem, 15-IV-1968, same host; 8 larvae (RML 54051), same locality and host, 11-VII-1970; 4 larvae (RML 56189), same locality and host, 5-VIII-1970; 1 larva (RML 50352), APEG Forest, Belem, same host, 22-X-1968, 3 larvae (RML 51728), same locality and host, 29-IV-1968 Thomas H. G. Aitken coll.;

2 larvae (RML 56187), same data; 5 larvae (RML 56188), same locality and host, 4-VIII-1970; 2 larvae (RML 54061), same locality and host, 9-VI-1970; 1 larva (RML 54052), same locality and host, 14-VIII-1970; 9 larvae (RML 54058), same locality and host, 24-IV-1970; 5 larvae (RML 56182), same locality and host, 22-VII-1970; 1 larva (RML 56183), same data; 2 larvae (RML 50353), same locality, 29-X-1968, *Proechimys guyannensis*; 8 larvae (RML 56186), same locality and host, 17-VII-1970; 1 larva (RML 56191), same locality and host, 6-VIII-1970; SURINAM – 1 larva (RML 49990), Paloemeu Airstrip, Tapanahoni river, 30-V-1961, *Hylaeamys laticeps*, H. A. Beatty coll.

**Distribution (Fig. 4).** Brazil - Amapá state: Serra do navio municipality, ICOMI [Indústria e Comércio de Minérios S.A.] ( $0^{\circ} 54' 23.393''$  N,  $51^{\circ} 59' 57.977''$  W); Pará state: Belém municipality, APEG [Área de Pesquisa Ecológica do rio Guamá/ Ecological Research of rio Guamá Area] ( $1^{\circ} 27' 12.672''$  S,  $48^{\circ} 25' 1.726''$  W); Catu Forest ( $1^{\circ} 26' 23.838''$  S,  $48^{\circ} 26' 29.029''$  W); Bragança municipality, Caratateua ( $0^{\circ} 59' 28.604''$  S,  $46^{\circ} 43' 19.301''$  W) (Brennan 1970). Venezuela – Bolívar state: Icabarú city ( $4^{\circ} 19' 22.854''$  N,  $61^{\circ} 45' 4.108''$  W) (Brennan & Reed 1975). Surinam - Paloemeu Airstrip, Tapanahoni River ( $3^{\circ} 20' 44.999''$  N,  $55^{\circ} 26' 30.001''$  W) (Brennan 1970); Moederzorg district: Babunhol ( $5^{\circ} 13' 27.091''$  N,  $55^{\circ} 1' 59.423''$  W); Brokopondo district: Brownsberg Nature Park ( $4^{\circ} 55' 59.999''$  N,  $55^{\circ} 10' 59.999''$  W); Saramacca district: Santo Boma ( $5^{\circ} 46' 0.001''$  N,  $55^{\circ} 34' 59.999''$  W) (Brennan & van Bronswijk 1975).

**Hosts.** Rodentia - Cricetidae: *Euryoryzomys macconnelli* (Thomas, 1910), *Hylaeamys laticeps* (Lund, 1840) and *Hylaeamys megacephalus* Fischer, 1814 (= *Oryzomys capito*); Dasyprotidae: *Myoprocta acouchy* (Erxleben, 1777); Echimyidae: *Proechimys guyannensis* (Geoffroy, 1803).

***Herpetacarus (Herpetacarus) hertigi* (Brennan & Jones, 1964) (Figs. 2 and 3)**

*Euschoengastia hertigi* Brennan and Jones 1964: 308.

*Herpetacarus (Abonnencia) hertigi*: Vercammen-Grandjean 1966: 642.

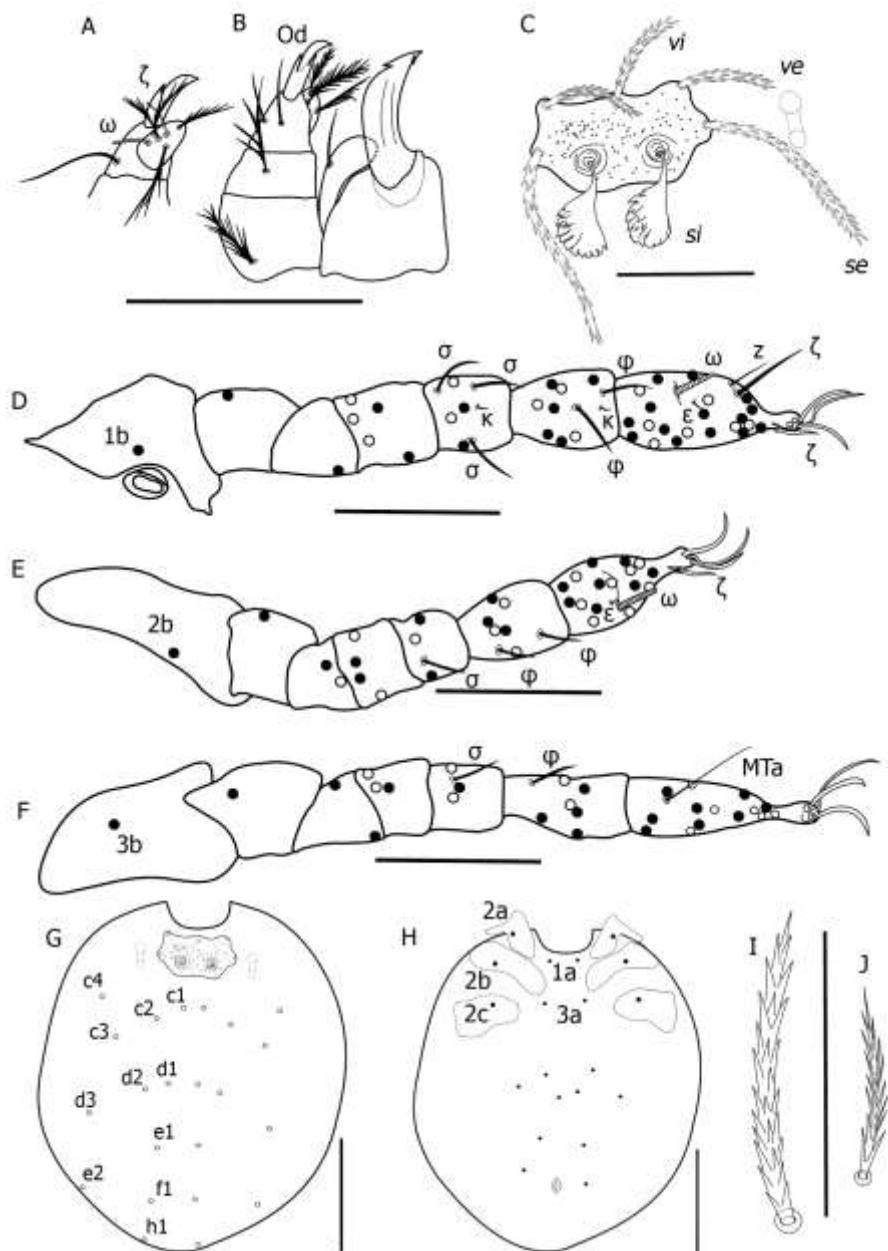
*Arisocerus hertigi*: Brennan 1970: 32; Whitaker and Dietz 1987: 191; Goff and Gettinger 1989: 557; Regolin *et al.* 2015: 38; Jacinavicius *et al.* 2018a: 6; Jacinavicius *et al.* 2018b: 208.

*Herpetacarus (Herpetacarus) hertigi* com. nov.

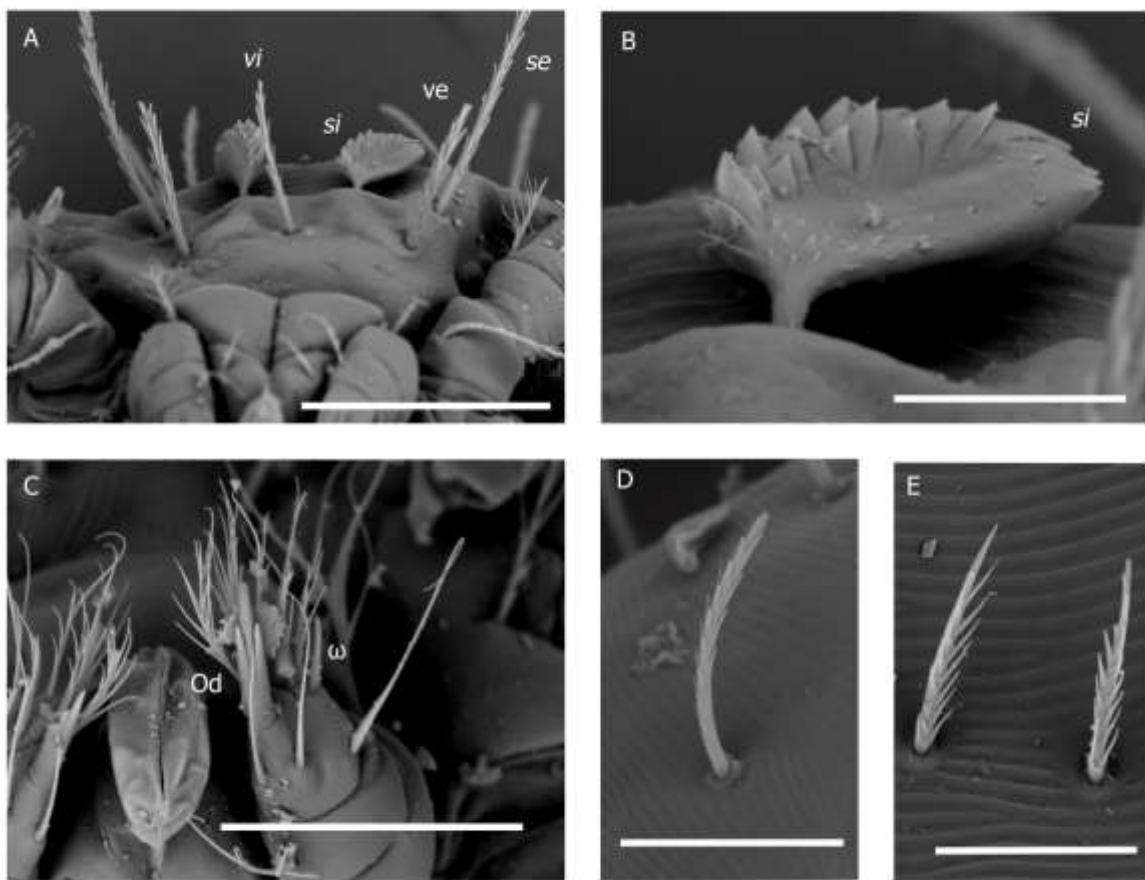
**Diagnosis.** Larva. Palptibia with the dorsal, lateral and ventral nude setae; adoral setae nude; idiosoma with 36 setae; setae *si* expanded asymmetrical, tarsus of the legs I with  $\epsilon$  distal to  $\omega$ , tarsus of the leg II with  $\epsilon$  proximal to  $\omega$ , tarsus of the legs III with mastisetae.

**Redescription (Holotype and Paratypes).** Larvae. *Gnathosoma* - palp setal formula B/B/NNN/7B $\zeta\omega$ ; odontus trifurcate, supracoxal setae branched, cheliceral blade with tricuspid cap; gnathobase punctate, adoral setae nude (Figs. 2A-B, 3C). *Idiosoma* - eyes 2/2 set in an ocular plate, anterior eye larger than the posterior eye; prodorsal sclerite with 1 pair of expanded assymetrical internal scapular trichobothridial setae (*si*) with short and robust setules, a single *vi* (= AM) seta, 1 pair of *ve* (= AL) seta and one pair of *se* (= PL) seta on the prodorsal sclerite; *se* > *si* > *vi* > *ve*; the anterior and posterior margin of the prodorsal sclerite undulating, and the lateral margins slightly concave (Fig. 2C, 3A-B-). Idiosoma with 36 setae, 4 pairs of setae in the C row, and the  $c_4$  pair of seta in an anterior position (= humeral setae), D row with 3 pairs of setae, E row with 2 pairs of setae, F row with 1 pair of setae, H row with 1 pair of setae, totalizing 22 dorsal opisthosomal setae, 2 pairs of sternal setae (1a, 3a), and 10 setae located anterior of the anus, totalizing 14 ventral setae (Figs. 2G-J, 3D-E). Legs - femur legs I-III each divided into a basifemur and telofemur, each leg terminating with a pair of claws and a claw like empodium, without onychotriches, coxal fields not striate. *Leg I* - coxal field with 1 branched seta *1b* (1B); trochanter 1B; basifemur 1B; telofemur 5B; genu 4B, 3  $\sigma$ , with  $\kappa$ ; tibia 8B, 2  $\varphi$ , with  $\kappa$ ; tarsus 21B, with  $\omega$ ,  $\epsilon$ , dorsal eupathid ( $\zeta$ ) with a companion seta (*z*) and terminating with a subterminal eupathid ( $\zeta$ ), famulus ( $\epsilon$ ) distal to  $\omega$  (Fig. 2D). *Leg II* - coxal field seta *2b* (1B); trochanter 1B; basifemur 2B; telofemur 4B; genu 3B,  $\sigma$ ; tibia 6B, 2  $\varphi$ ; tarsus 15B, with  $\omega$ ,  $\epsilon$ , and a subterminal eupathid ( $\zeta$ ), base of  $\epsilon$  proximal to  $\omega$  (Fig. 2E). *Leg III* - coxal field seta *3b* (1B), trochanter 1B; basifemur 2B; telofemur 3B; genu 3B,  $\sigma$ ; tibia 6B,  $\varphi$ ; tarsus 14B; 1 mastisetae (Fig. 2F).

**Remarks.** In the original description the author reposted 12 setae on the ventral side of the idiosoma, reexamining the three paratypes we observed 14 setae.



**Figure 2.** Morphological details of *Herpetacarus (Herpetacarus) hertigi* com. nov. larva. A. ventral view of palptarsus; B. dorsal view of gnathosoma; C. prodorsal sclerite; D. Leg I; E. Leg II; F. Leg III; G. dorsal view of idiosoma; H ventral view of idiosoma; I. dorsal opisthosomal setae; J. ventral opisthosomal setae. Symbols: ve = external vertical setae; se = external scapular setae; vi = internal vertical setae; si = internal scapular setae or trichobothria;  $\omega$  = solenidion of palptarsus and of the tarsus on the leg I and II; Od = odontus;  $\zeta$  = dorsal eupathid of the tarsus of the leg I and subterminal eupathid of the tarsus of the leg I and II and of the palptarsus;  $\sigma$  = solenidia on the genu of the legs I, II and III;  $\kappa$  = microsetae on genu and tibia of the leg I;  $\varphi$  = solenidia on the tibia of the legs I, II and III;  $\varepsilon$  = famulus on the tarsus of the legs I and II;  $z$  = companion seta on tarsus of the leg I; MTa = Marstiseta on the tarsus of the leg III; 2b = seta of the coxal field of the leg II; 3b = seta of the coxal field of the leg III; c<sub>1</sub>-c<sub>4</sub> = setae of the C row; d<sub>1</sub>-d<sub>3</sub> = setae of the D row; e<sub>1</sub>-e<sub>2</sub> = setae of the E row; f<sub>1</sub> = setae of the F row; h<sub>1</sub> = setae of the H row; 1a = anterior sternal setae; 3a = posterior sternal setae; Black spots = ventral setae of the idiosoma; white spots = dorsal setae of the idiosoma. Scales: A-F 50µm; G and H 100 µm; I and J 50µm.

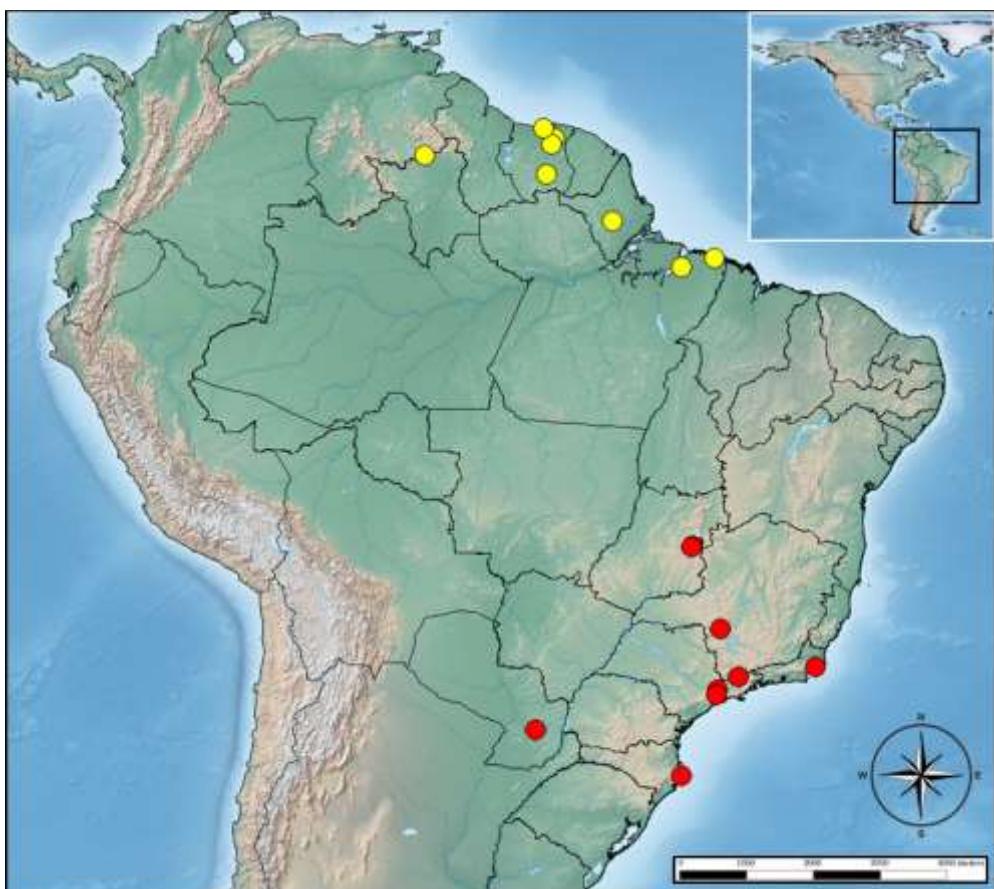


**Figure 3.** Scanning electron micrographs of *Herpetacarus (Herpetacarus) hertigi* **com. nov.** larva. A. prodorsal sclerite; B. internal scapular setae or trichobothria; C. dorsal view of gnathosoma; D. dorsal opisthosomal setae; E. ventral opisthosomal setae. Symbols: *ve* = external vertical setae; *se* = external scapular setae; *vi* = internal vertical setae; *si* = internal scapular setae or trichobothria;  $\omega$  = solenidion of palptarsus; *Od* = odontus. Scales: A 50 $\mu$ m; B 10  $\mu$ m; C and D 30 $\mu$ m; E 20  $\mu$ m.

**Type and material examined.** PARATYPES: 16 larvae (RML 43427), Sommerfeld [Sommerfeld Colony, Caaguazú Department], Paraguay, 3-III-1950, “agouti” [*Dasyprocta* sp.]; 8 larva (RML 44504), same locality, 11-III-1950, “opossum” [*Didelphis* sp.]. Material: BRAZIL - 6 larvae (IBSP 11989A), The Moleques do Sul Archipelago, Santa Catarina, 10-X-2009, *Cavia intermedia*; 2 larvae (IBSP 11990), same locality and host, 11-X-2009; 3 larvae (IBSP 11991A), same locality and host, 13-X-2009; 4 larvae (IBSP 10518), same locality and host, 10-18-IX-2008; 1 larva (IBSP 1155), Serra da Cantareira, São Paulo, São Paulo state, 13-IX-193, *Cuniculus paca*; 4 larvae (IBSP 10519), Zoológico de São Paulo, São Paulo, São Paulo state, 16-IV-2010, *Oligoryzomys* sp.; 10 larvae (IBSP 10520), same data; 1 larva (IBSP 10521B), same locality and date, *Akodon* sp.; 10 larvae (IBSP 10389), same locality and date, *Akodon* sp; 1 larvae (IBSP 11239), Campos do Jordão, São Paulo state, 13-I-2013, *Akodon*

*montensis*; 3 larvae (IBSP 13068), Fazenda Agua Limpa, Brasilia, Federal District, 22-IV-1984, *Hylaeamys megacephalus*.

**Distribution (Fig. 4).** Brazil – Brasilia, Federal district ( $47^{\circ} 52' 46.772''$  W,  $15^{\circ} 56' 45.294''$  S) (Goff & Gettinger 1989); Fazenda Agua Limpa ( $47^{\circ} 56' 00.8''$  W,  $15^{\circ} 56' 55.6''$  S) (This study); Minas Gerais state: Fazenda das Pedras, Serra da Canastra national Park ( $46^{\circ} 24' 10.141''$  W,  $20^{\circ} 14' 42.630''$  S) (Whitaker & Dietz 1987); Rio de Janeiro state: Restinga National Park of Jurubatiba ( $41^{\circ} 29' 33''$  W  $22^{\circ} 12' 5''$  S) (Jacinavicius, 2018b); Santa Catarina state: Moleques do Sul Arquipelago ( $48^{\circ} 25' 54.088''$  W,  $27^{\circ} 50' 47.180''$  S) (Regolin et al. 2015); São Paulo state: Campos do Jordão municipality ( $45^{\circ} 28' 53.893''$  W,  $22^{\circ} 41' 24.025''$  S) (This study); Serra da Cantareira National Park ( $46^{\circ} 35' 5.197''$  W,  $23^{\circ} 24' 19.130''$  S) (This study); Zoológico de São Paulo ( $46^{\circ} 37' 13.490''$  W,  $23^{\circ} 38' 42.292''$  S) (This study). Paraguay - Caaguazú Department: Sommerfeld Colony (Brennan & Jones 1964).



**Figure 4.** Geographical distribution of *Herpetacarus (Herpetacarus) amapensis* com. nov. (yellow spots) and *Herpetacarus (Herpetacarus) hertigi* com. nov. (red spots).

**Hosts.** Didelphimorphia – Didelphidae: *Didelphis albiventris* Lund, 1840 and “opossum” [*Didelphis* sp.]; Rodentia – Caviidae: *Cavia intermedia* Cherem, Olimpio & Ximenez, 1999; Cuniculidae: *Cuniculus paca* (Linnaeus, 1766); Dasyprotidae: “Agouti” [*Dasyprocta* sp.]; Cricetidae: *Akodon* sp., *Akodon montensis* Thomas, 1913, *Hylaeamys megacephalus* Fischer, 1814, *Nectomys squamipes* (Brants, 1827), *Oligorysomys fornesi* (Massoia, 1973), *Oligoryzomys* sp. and *Oxymycterus* sp.

#### 6.4 DISCUSSION

Regarding of the examination, depending on the position of the mite on the slide, the quantity of branched setae on the palptarsus and the presence or absence of an euphatid ( $\zeta$ ) is not clearly visible and may cause errors of identification. Brennan and Jones (1964), in the description of *H. hertigi* **com. nov.**, were not sure if the palptarsus has 6 or 7 branched setae (6B $\omega$  or 7B $\omega$ ). Vercammen-Grandjeanm (1966) in the review of *Herpetacarus* consider *H. hertigi* **com. nov.** of the subgenus *Abonnencia* and described this species with 7B $\omega$  on the palptarsus. Brennan (1970) described a genus *Arisocerus* with 6B $\omega\zeta$  on the palptarsus and included the species *H. hertigi* **com. nov.** and *H. amapensis* **com. nov.**. Goff & Gettinger (1989) reexamined the types of the both species, and observed the palptarsus having 7B $\omega\zeta$ , and still considered the genus *Arisocerus* as valid by the shape of trichobrthrias. We confirm this character (7B $\omega\zeta$ ), futhermor, based in the morphological characters of the both species examined, the validity of the genus *Arisocerus* is in question because the diagnosis is identical to the genus *Herpetacarus* (*Herpetacarus*). Here, we are considered *Arisocerus* is a junior synonym of *Herpetacarus* (*Herpetacarus*). Besides that, originally the genus *Herpetacarus* was created to group species collected parasitizing reptiles, but later works showed some species parasitizing vertebrates of other order, including mammals.

*Herpetacarus* (*Herpetacarus*) *amapensis* **com. nov.** was recorded in Venezuela, Surinam and north region of Brazil associated with rodents of family Dasyprotidae, Echimyidae and Cricetidae (Brennan 1970; Brennan & Reed 1975; Brennan & van Bronswijk 1975). The species *H. hertigi* **com. nov.** have been reported in Paraguay and southern, southeastern and central regions of Brazil associated with marsupials of the family Didelphidae and rodents of the family Caviidae and Cricetidae (Brennan & Jones 1964; Goff & Gettinger 1989; Regolin *et al.* 2015; Whitaker & Dietz 1987). In the present study we reported the first record of *H. hertigi* **com. nov.** in the São Paulo and Campos do Jordão

municipality, São Paulo state and the first record of the parasitism in the ears of the rodents *Akodon montensis*, *Cuninulus paca* and *Hylaeamys megacephalus*.

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## 7. CHAPTER 6 – NEW RECORDS OF CHIGGERS PARASITIZING MAMMALS IN BRAZIL HOUSED AT NATIONAL AND INTERNATIONAL ACARI COLLECTIONS

### 7.1 INTRODUCTION

Currently, there are more than 3700 chigger mites species recorded worldwide parasitizing vertebrates of the classes Amphibia, Reptile, Aves and Mammals and are organized in three families: Trombiculidae Ewing, 1944, Leeuwenhoekiidae Womersley, 1944 and Walchiidae Ewing, 1946 *sensu* Wen, 1999 (Zhang, 2011). According to the IUCN (2019) are known 3688 native species of vertebrates in Brazil, of these 641 are terrestrial species belongs to the Class Mammalia. However, only 43 species of the orders Cetartiodactyla, Chiroptera, Didelphimorphia, Pilosa, Primates and Rodentia were found parasitized by chiggers in Brazil (Bassini-Silva et al., 2018; Jacinavicius et al., 2018a, b, c 2019 [in press]).

Institution that housed natural history collections are important biodiversity repositories and were consulted and the material of chiggers parasitizing mammals were examined and listed below. Besides that, we add new records of locality and hosts for chiggers in Brazil.

### 7.2 MATERIAL AND METHODS

Brazilian and American institutions that housed collections of mites were consulted, and those that had specimens of chiggers collected in Brazil parasitizing mammals. The following institutions were consulted: Coleção Acarológica do Instituto Butantan (IBSP), São Paulo, Brazil; Museu de Zoologia da USP (MZUSP), São Paulo, Brazil; Instituto Oswaldo Cruz (CAVAISC-IOC), Rio de Janeiro, Brazil; Coleção Científica de Acari UNESP (DZSJRP-ACARI), São José do Rio Preto, Brazil; Coleção de Ácaros da ESALQ USP (AcariESALQ), Piracicaba, Brazil; Coleção do Instituto Biológico (IB), São Paulo, Brazil; Museu de História Natural Capão da Imbuia (MHNCI), Curitiba, Brazil; Museu Paraense Emílio Goeldi (MPEG), Belém, Brazil, Instituto Nacional de Pesquisas da Amazonia (INPA), Amazonas, Brazil; Acari Collection from National Museum of Natural History (NMNH), Maryland, USA and Ohio State Acarological Laboratory (OSAL), Ohio, USA.

The material, previously preserved in alcohol, was slide-mounted in Hoyer's medium, according Walter and Krantz (2009), while specimens preserved in old slides were recovered according to Jacinavicius et al. (2013). Posteriorly, all the material was identified at the genus level using a key published by Brennan and Goff (1977). Subsequently, specimens were identified to the species level by comparing them to the original description of all species of the genus.

### 7.3 RESULTS

The following collections consulted have samples of chiggers collected parasitizing mammals from Brazil: CAVAIS-IOC, NMNH, OSAL, DZSJR-ACARI and IBSP. In the other institutions do not have in their collections type or material of chiggers that parasitize mammals. Here, we are including new records of localities and hosts of chiggers recently examined from the IBSP and DZSJR-ACARI collections. A total of 224 chigger mites were examined to the family Trombiculidae were listed below.

#### **TROMBICULIDAE Ewing, 1944**

***Boshkerria punctata* (Bosshell and Kerr, 1942):** 1 larva (IBSP 12486B), Fazenda Nhumirim, Nhecolândia, Mato Grosso do Sul State, Brazil, 04-III-2015, *Thrichomys fosteri* (Rodentia).

***Eutrombicula alfreddugesi* (Oudemans, 1910);** 1 larva (IBSP 11056A), Inhamum, Caxias, Maranhão State, Brazil, 01-VII-2011, *Didelphis aurita* (Didelphimorphia); 1 larva (IBSP 12487C), Fazenda Nhumirim, Nhecolândia, Mato Grosso do Sul State, Brazil, 04-III-2015, *Thrichomys fosteri* (Rodentia); 5 larvae (IBSP 12090A), Serra das Confusões, Piauí State, Brazil, 24-V-2013, *Thricomys apereroides* (Rodentia).

***Eutrombicula batatas* (Linnaeus, 1758);** 8 larvae (IBSP 341) Butantan, São Paulo municipality, São Paulo State, Brazil, 26-X-1933, *Didelphis aurita* (Didelphimorphia).

***Eutrombicula goeldii* (Oudemans, 1910);** 3 larvae (IBSP 11056B), Inhamum, Caxias, Maranhão State, Brazil, 01-VII-2011, *Didelphis aurita* (Didelphimorphia); 2 larvae (IBSP 11054), same locality and data, *Marmosa murina* (Didelphimorphia); 2 larvae (IBSP

11053A), same locality, 30-VII-2011, *Marmosa demerarae* (Didelphimorphia). 1 larva (IBSP 908), Anápolis municipality, Goiás State, Brazil, 10-X-1936, Rodentia.

***Eutrombicula tinami* (Oudemans, 1910):** 2 larvae (IBSP 352), Butantan, São Paulo municipality, São Paulo State, Brazil, 24-V-1932, *Didelphis aurita* (Didelphimorphia); 1 larva (IBSP 1680), Vila Albertina, São Paulo municipality, São Paulo State, Brazil, 25-II-1938, *Didelphis albiventris* (Didelphimorphia); 4 larvae (IBSP 10541), Barra do Una, Peruíbe municipality, São Paulo State, Brazil, 05-XI-2010, *Didelphis aurita* (Didelphimorphia); 1 larva (IBSP 11255), Condomínio Vila Verde, Itapevi municipality, São Paulo State, Brazil, 13-XII-2012, same host; 1 larva (IBSP 10535C), Parque Estadual Cantareira, São Paulo municipality, São Paulo State, Brazil, 12-IV-2011, *Euryoryzomys russatus* (Rodentia); 4 larvae (IBSP 1654), Parque do Estado da Água Funda, São Paulo municipality, São Paulo State, Brazil, 07-III-1939, *Hidrochoerus hidrochoeris* (Rodentia); 2 larvae (IBSP 10549), Confresa municipality, Mato Grosso State, Brazil, 26-IV-2011, *Didelphis marsupialis* (Didelphimorphia); 6 larvae (IBSP 10551), same locality and host, 29-IV-2011; 1 larva (IBSP 358), Angra dos Reis municipality, Rio de Janeiro State, Brazil, VI-1931, *Dasyprocta leporina* (Rodentia); 1 larva (IBSP 1681), Abaeté municipality, Minas Gerais State, Brazil, 05-VII-1937, *Proechimys roberti* (Rodentia); 2 larvae (IBSP 1684), same locality and host, 01-VI-1937; 1 larva (IBSP 5754), Flores, Manaus municipality, Amazonas State, Brasil, 08-VIII-1955, *Dasyprocta* sp. (Rodentia); 4 larvae (IBSP 353), Butantan, São Paulo municipality, São Paulo State, 21-V-1932, *Sylvilagus brasiliensis* (Lagomorpha); 1 larva (IBSP 1975), Serra da Cantareira, São Paulo municipality, São Paulo State, Brazil, 12-VIII-1940, *Nasua narica* (Carnivora).

***Hooperella vesperuginis* (Brennan and Jones, 1960):** 1 larva (IBSP 2127D), Butantan, São Paulo municipality, São Paulo State, Brazil, 29/V/1954, *Glossophaga soricina* (Chiroptera).

***Microtrombicula brachytrichia* Brennan, 1971:** 2 larvae (IBSP 13089), Japecanga, Parque Nacional da Serra das Confusões, Piauí State, Brazil, 02-VIII-2013, *Monodelphis domestica* (Didelphimorphia); 1 larva (IBSP 12092A), Serra das Confusões, Piauí State, Brazil, 24-V-2013, *Rhipidomys* sp. (Rodentia).

***Microtrombicula brennani* Goff, Whitaker and Dietz, 1986:** 1 larva (IBSP 11715), Anastásio municipality, Mato Grosso do Sul State, Brazil, 21-II-2012, *Didelphis albiventris* (Didelphimorphia).

***Microtrombicula rhipidomysi* Goff, Whitaker and Dietz, 1983:** 1 larva (IBSP 10526C), Barra do Una, Peruíbe municipality, São Paulo State, Brazil, 16-VI-2010, *Euryoryzomys russatus* (Rodentia); 1 larva (IBSP 11701), Terenos municipality, Mato Grosso do Sul State, Brazil, 22-I-2012, *Thylamys macrurus* (Rodentia); 1 larva (IBSP 11702), Miranda municipality, Mato Grosso do Sul State, Brazil, 11-II-2012, *Thylamys macrurus* (Rodentia); 2 larvae (IBSP 11704), Bonito municipality, Mato Grosso do Sul State, Brazil, 1-III-2012, same host; 2 larvae (IBSP 11721), same data; 1 larva (IBSP 11726), same locality and host, 17-VI-2013; 1 larva (IBSP 11727A), same locality and host, 2-III-2012; 1 larva (IBSP 11699), same locality and host, 3-VI-2014; 1 larva (IBSP 11728), Miranda municipality, Mato Grosso do Sul State, Brazil, 8-II-2012, same host; 1 larva (IBSP 11729), same locality and host, 09-II-2012; 1 larva (IBSP 11730), same locality and host, 13-II-2012; 2 larvae (IBSP 12091E), Serra das Confusões, Piauí State, Brazil, 24-V-2013, *Rhipidomys* sp. (Rodentia). 1 larva (IBSP 1099), Serra da Cantareira, São Paulo municipality, São Paulo State, 03-VI-1937, *Puma concolor* (Carnivora); 1 larva (IBSP 13095A), Canto Verde, Parque Nacional da Serra das Confusões, Piauí State, Brazil, 22-X-2013, *Kerodon rupestris* (Rodentia).

***Parascoschoengastia aemulata* (Brennan and Jones, 1964a):** 2 larvae (IBSP 12041C), Serra das Confusões, Piauí State, Brazil, 22-V-2013, *Thricomys apereroides* (Rodentia).

***Parasecia valida* Brennan, 1969b:** 1 larva (IBSP 12041D), Serra das Confusões, Piauí, Brasil, 22-V-2013, *Thricomys apereroides* (Rodentia).

***Perissopalla tanydera* Brennan, 1969a:** 1 larva (DZSJRP-ACARI 2086), Fazenda Lageado, Botucatu municipality, São Paulo State, Brazil, 29-VI-1995, *Micronycteris* (*Neonycteris*) *pusilla* (Chiroptera).

***Quadrasetia azulae* (Brennan and Jones, 1964b):** 1 larva (IBSP 1932), Butantan, São Paulo municipality, São Paulo State, Brazil, 19-IV-1940, *Akodon cursor* (Rodentia); 2 larvae (IBSP 343), Serra da Cantareira, São Paulo State, Brazil, 1-VIII-1935, *Guerlinguetus ingrami* (Rodentia); 2 larvae (IBSP 1700), same locality, 29-VII-1939, *Didelphis aurita*

(Didelphimorphia); 1 larva (IBSP 382A), Butantan, São Paulo municipality, São Paulo State, Brazil, 23-X-1933, *Sylvilagus brasiliensis* (Lagomorpha);

***Quadraseta brasiliensis* Goff and Gettinger, 1989:** 2 larvae (IBSP 11110E), Morro Grande, Cotia municipality, São Paulo State, Brazil, 16-VII-2012, *Nectomys squamipes* (Rodentia); 7 larvae (IBSP 12706B), Morro Alto, São Francisco do Sul municipality, Santa Catarina State, Brazil, 19-V-2013, same host; 1 larva (IBSP 12707B), same data; 2 larvae (IBSP 12708B), same data; 1 larva (IBSP 12709B), same data; 2 larvae (IBSP 12713), same locality and host, 22-V-2013; 3 larvae (IBSP 12710A), same locality, 20-V-2013, *Akodon* sp. (Rodentia); 2 larvae (IBSP 12712), same locality and date, *Oligoryzomys nigripes* (Rodentia); 1 larva (IBSP 11718A), Bonito, Mato Grosso do Sul State, Brazil, 12-III-2012, *Hylaeamys megacephalus* (Rodentia); 2 larvae (IBSP 11719A), same locality and host, 03-VI-2013; 2 larvae (IBSP 11722B), same locality and host, 06-VI-2013; 1 larva (IBSP 11725A), same locality and host, 09-III-2012; 4 larvae (IBSP 13070), Fazenda Água Limpa, Brasília, Federal District, Brazil, 17-III-1984, *Necromys lasiurus* (Rodentia).

***Quadraseta falconensis* Goff and Brennan, 1977:** 7 larvae (IBSP 11724), Terenos municipality, Mato Grosso do Sul State, Brasil, 13-VII-2013, *Gracilinamus agilis* (Didelphimorphia); 1 larva (IBSP 11700), Miranda municipality, Mato Grosso do Sul State, Brazil, 09-II-2012, *Thylamys macrurus* (Rodentia); 1 larva (IBSP 11703), Bonito municipality, Mato Grosso do Sul State, Brazil, 11-III-2012, same host.

***Quadraseta flochi* (Brennan and Jones, 1960):** 2 larvae (IBSP 11055), Inhamum, Caxias municipality, Maranhão State, Brazil, 15-VII-2010, *Didelphis albiventris* (Didelphimorphia); 2 larvae (IBSP 11053B), same locality, 30-VII-2011, *Marmosa demerarae* (Didelphimorphia); 1 larva (IBSP 12039), Parque Nacional da Serra das Confusões, Piauí State, Brazil, 23-I-2013, *Monodelphis domestica* (Didelphimorphia); 6 larvae (IBSP 13082), same locality and host, 30-VII-2013; 1 larva (IBSP 13092A), same locality, 02-VIII-2013, *Thrichomys apereroides* (Rodentia); 1 larva (IBSP 13113B), same locality and host, 24-X-2013; 2 larvae (IBSP 11539B) Anastácio municipality, Mato Grosso do Sul State, Brazil, 20-II-2012, *Rhipidomys macrurus* (Rodentia).

***Quadraseta mackenziei* (Yunker and Brennan, 1964):** 1 larva (IBSP 11723A), Bonito municipality, Mato Grosso do Sul State, Brazil, 02-III-2012, *Monodelphis domestica*

(Didelphimorphia); 6 larvae (IBSP 11720A), same locality, 07-VI-2013, *Hylaeamys megacephalus* (Rodentia).

***Quadraseta pazca* (Brennan and Jones, 1964b):** 4 larvae (IBSP 345), Campinas municipality, São Paulo State, Brazil, 1-III-1935, *Didelphis albiventris* (Didelphimorphia); 1 larva (IBSP 11241A), Parque Estadual dos Mananciais, Campos do Jordão municipality, São Paulo State, Brazil, 13-I-2013, *Akodon* sp. (Rodentia); 4 larvae (IBSP 10535B), Parque Estadual Cantareira, São Paulo municipality, São Paulo State, Brazil, 12-IV-2011, *Euryoryzomys russatus* (Rodentia); 1 larva (IBSP 11367B), Sete Barras municipality, São Paulo State, Brazil, 15-VIII-2013, *Oxymycterus* sp. (Rodentia); 3 larvae (IBSP 11365B), same locality, 10-VIII-2013, *Euryoryzomys russatus* (Rodentia); 3 larvae (IBSP 10522B), Barra do Una, Peruíbe municipality, São Paulo State, Brazil, 08-XI-2010, *Euryoryzomys russatus* (Rodentia); 2 larvae (IBSP 10523), same locality and host, 16-IV-2010; 1 larva (IBSP 10524B), same locality and host, 18-VI-2010; 1 larva (IBSP 10525B), same data; 2 larvae (IBSP 10526A), same locality and host, 16-VI-2010; 1 larva (IBSP 10527B), same data; 2 larvae (IBSP 10530B), same locality and host; 20-IV-2010; 1 larva (IBSP 10531B), V, 21-IV-2010; 3 larvae (IBSP 10533B), V, 19-IV-2010; 1 larva (IBSP 12710B), Morro Alto, São Francisco do Sul municipality, Santa Catarina State, Brazil, 20-V-2013, same host; 14 larvae (IBSP 12706A), same locality and host, 19-V-2013; 1 larva (IBSP 12707A), same data; 1 larva (IBSP 12708A), same data; 1 larva (IBSP 12709A), same data; 3 larvae (IBSP 12711), same data; 1 larva (IBSP 12554B), Morro Grande, Cotia municipality, São Paulo State, Brazil, 17-VI-2015, *Thaptomys nigrita* (Rodentia); 1 larva (IBSP 12714), Morro Alto, São Francisco do Sul municipality, Santa Catarina State, Brazil, 19-V-2013, “wild rodent” (Rodentia).

***Quadraseta trapezoides* (Brennan and Jones, 1964b):** 2 larvae (IBSP 1705), Serra da Cantareira, São Paulo State, Brazil, 03-VIII-1939, “wild rodent” (Rodentia); 7 larvae (IBSP 12706B), Morro Alto, São Francisco do Sul municipality, Santa Catarina State, Brazil, 19-V-2013, *Nectomys squamipes*.

***Serratacarus dietzi* Goff and Whitaker, 1984:** 2 larvae (IBSP 1122), Serra da Cantareira, São Paulo State, Brazil, 02-VI-1937, “cuica” (Didelphimorphia).

***Speleocola tamarina* Goff, Whitaker and Dietz, 1987:** 1 larva (IBSP 11698C), Bonito municipality, Mato Grosso do Sul State, Brazil, 07-VII-2012, *Hylaeamys megacephalus* (Rodentia); 1 larva (IBSP 11540B), Anastácio municipality, Mato Grosso do Sul State, Brazil, 21-II-2012, *Rhipidomys macrurus* (Rodentia); 1 larva (IBSP 11549B), Bonito municipality, Mato Grosso do Sul State, Brazil, 10-III-2012, same host.

***Trombewingia bakeri* (Fonseca, 1955):** 2 larvae (IBSP 12143A), Morro Grande, Cotia municipality, São Paulo State, Brazil, 15-IV-2015, *Akodon montensis* (Rodentia).

#### 7.4 DISCUSSION

The material and types from chiggers housed at the collection CAVAIS-IOC have already been published by Jacinavicius et al (2018a, b), as well as, the collection of chiggers housed in NMNH and OSAL, constituted mainly by primary and secondary types, were summarized in a checklist by Jacinavicius et al. (2018a).

The examination of material deposited in collections was fundamental to increase the number of records and distribution of chiggers in Brazil. In this way it was possible to access material from different dates and localities. In this study, is being recorded the first report of chiggers in the state of Piauí, besides that, more records for several localities of the states of Amazonas, Goiás, Maranhão, Mato Grosso, Mato Grosso do Sul, Minas Gerais, Rio de Janeiro, Santa Catarina, São Paulo, have been recorded here.

In addition, this is the first report of chiggers parasitizing hosts of the orders Carnivora (*Nasua narica* and *Puma concolor*) and Lagomorpha (*Sylvilagus brasiliensis*) in Brazil. Besides that, the others orders, in this study, has been already recorded, however the species *Glossophaga soricina* (Chiroptera), *Micronycteris (Neonycteris) pusilla* (Chiroptera), *Marmosa demerarae* (Didelphimorphia), *Marmosa murina* (Didelphimorphia), *Kerodon rupestris* (Rodentia), *Akodon cursor* (Rodentia), *Proechimys roberti* (Rodentia), *Thaptomys nigrita* (Rodentia), *Thricomys apereroides* (Rodentia) and *Thrichomys fosteri* (Rodentia) are also found parasites for the first time in Brazil by chiggers.

Regards of the chiggers, the genus *Boshkerria* Fauran 1959 is reported for the first time in Brazil, and the species *Boshkerria punctata*, *Quadraseta azulae*, *Quadraseta mackenziei* and *Hooperella vesperuginis*, increase the records of the species in Brazil, totalizing 73 species for the Brazilian territory. In addition, the species *Eutrombicula alfreddugesi*, *Eutrombicula batatas*, *Eutrombicula goeldii*, *Eutrombicula tinami*,

*Microtrombicula brachytrichia*, *Microtrombicula brennani*, *Microtrombicula rhipidomysi*, *Parascoschoengastia aemulata*, *Parasacia valida*, *Perissopalla tanydera*, *Quadraseta brasiliensis*, *Quadraseta falconensis*, *Quadraseta flochi*, *Quadraseta pazca*, *Quadraseta trapezoides*, *Speleocola tamarina* and *Serratacarus dietzi* has been new records of localities for Brazil.

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## 8. CHAPTER 7 – MOLECULAR DETECTION OF PATHOGENS IN CHIGGER MITES (TROMBIDIFORMES: TROMBICULIDAE) COLLECTED IN SMALL BRAZILIAN MAMMALS

### 8.1 INTRODUCTION

The chiggers (Trombidiformes: Trombiculidae) parasitize any terrestrial vertebrates in the larval stage, and during feeding, inject digestive enzymes into the skin and can cause severe skin reactions in the host. The skin reactions are popularly known as trombiculiasis and can transmit pathogens (EWING, 1944). In Asia-Pacific region, chiggers, known as tsutsugamushi (“tsutsuga” = disease, “mushi” = bug), are vectors of *Orientia tsutsugamushi*, that causing the Scrub typhus, an acute febrile disease in humans (RAPMUND et al., 1969, TAKAHASHI et al., 2004, KELLY et al., 2009; PHASOMKUSOLSIL et al., 2009). And recently,

Frank (1977) detected the presence of the protozoa *Hepatozoon erhardovae* in chigger of the species *Hirsutiella zachvatkini* (Schluger 1948), in Austria. Besides that, the bacterium *Coxiella burnetii*, that causing Q-fever, has already been detected in chiggers in Africa (DANIEL, 1961; KEPKA, 1965). Blanc et al. (1952), under the laboratory conditions, successfully described *C. burnetti* infection in the species *Neotrombicula autumnalis* (Shaw) and Le Gac et al. (1953) reported a Q-fever case in a laboratory technician who was manipulating chiggers collected from an endemic area in Equatorial Africa. *Rickettsia* spp. bacteria were detected in chiggers from Ukraine (VYSOTSKAYA and SLUGER, 1953), Japan (CHOI et al., 2007) and China (HUANG et al., 2017), as well as *Rickettsia monacensis* and *Rickettsia helvetica* in Slovakia (MITKOVA et al., 2015).

In Brazil, Fonseca (1932) emphasized the importance of the ectoparasite mites as potential vectors in the epidemiological cycle of rickettsial diseases. For the first time, Bassini-Silva et al. (2018a) detected *Rickettsia* sp. in *Blankaartia sinnamaryi* (Floch and Fauran, 1956) parasitizing bird.

Here, we are contributing with the knowledge of the molecular studies of chiggers parasitizing small mammals. In addition, we are reporting the molecular detection of two different species of *Rickettsia* in three species of chiggers collected from eight hosts.

## 8.2 MATERIAL AND METHODS

### 8.2.1 Molecular analyses

Chiggers preserved in ethyl alcohol PA stored in -80°C freezer, housed in the Acari Collection of the Instituto Butantan (IBSP) of São Paulo City, Brazil, were individually submitted to DNA extraction using the Guanidine Isothiocyanate (GT) lysis protocol following Chomczynski (1993). Each mite was placed in a plastic microtube, and punctured in the idiosomal region with a sterile needle (1.20 \* 40 - 18G). After DNA extraction, exoskeletons of the specimens were recovered and slide-mounted in order to perform a morphological identification according to Walter and Krantz (2009).

A conventional PCR targeting a partial fragment of the mite 18S ribosomal gene and a section of the mite mitochondrial cytochrome oxidase I (COI) gene were initially performed using primers listed in Table 1. All reactions included a positive (DNA extracted from the chigger species *B. sinnamaryi*) and a negative (DNA-free Milli-Q water) controls. Once 18S rRNA and COI gene-PCRs were used as endogenous control, the negative samples were excluded from further analyses.

To detect the presence of DNA of pathogens, the successfully extracted samples were submitted to PCR assays targeting gene portions of organism of the genera *Coxiella*, *Hepatozoon* and *Rickettsia* (Table 1).

Reactions included negative control (DNA-free Milli-Q water) and the appropriate positive control, which consisted of DNA extracted from *Coxiella*-infected *Ornithodoros* ticks, *Hepatozoon canis*-infected canine blood, or Vero cell-infected spotted fever group *Rickettsia* (*Rickettsia parkeri* strain NOD or *Rickettsia vini*). Reactions yielding amplicons of expected size were purified using the ExoSAP-IT (USB Corporation®) following the manufacturer instructions (3 µl of the ExoSAP e 7,5 µl of the DNA amplified). Sanger sequencing of the samples was performed at the “Centro de pesquisa sobre Genoma Humano e Células Tronco do Instituto de Biociências da USP”. Obtained sequences were assembled and primer trimmed with Geneious R9 (KEARSE et al. 2012), and then submitted to a BLAST analysis ([www.ncbi.nlm.nih.gov/blast](http://www.ncbi.nlm.nih.gov/blast)) in order to infer closest similarities with other homologous sequences (ALTSCHUL et al., 1990)

**Table 1.** Oligonucleotide primers for amplification of 18S and COI genes (endogenous control), *gltA* and *ompA* (*Rickettsia*), 16S (*Coxiella*) and 18S (*Hepatozoon*).

Organisms	Gene	Sequence 5'-3'	Size	Reference
<i>Rickettsia</i> spp.	<i>gltA</i>	CS-78 (GCAAGTATCGGTGAGGATGTAAT)	401-pb	LABRUNA et al., 2004
		CS-323 (GCTTCCTTAAAATTCAATAAACAGGAT)		
<i>Rickettsia</i> spp. (GFM)	<i>ompA</i> (1st PCR)	Rr190.70 (ATGGCGAATATTCTCCAAAA)	632-pb	REGNERY et al., 1991 and ROUX et al., 1996
		Rr190.701R2 (GTTCCGTTAATGGCAGCATCT)		
<i>Rickettsia</i> spp. (GFM)	<i>ompA</i> (hemi-nested)	Rr190.70 (ATGGCGAATATTCTCCAAAA)	532-pb	REGNERY et al., 1991
		Rr190.602 (AGTGCAGCATTGCTCCCCCT)		
<i>Coxiella</i> spp.	16S	16SrRNA F (GGGGAGAAAGTCTCAAGGGTAATATCCTT)	532-pb	ALMEIDA et al., 2012
		16SrRNA R (TGCATCGAATTAAACCACATGCTCCACCGC)		
<i>Hepatozoon</i> spp.	18S	HEP142-169-F (GCTTGAAACACTCTARTTTCTCAAAG)	574-pb	ALMEIDA et al., 2013
		HEP743-718-R (ACAATAAAGTAAAAAACAYTTCAAAG)		
Mites	COI	COI 772 (TGATTTTTGGTCACCCAGAAG)	408-pb	NAVAJAS (1994) adapted by SOLLER et al. (2001)
		COI 773 (TACAGCTCCTATAGATAAAAC)		
Mites	COI	bcdF01: CATTTCCHACTAACYCATARGATATTGG bcdR04: TATAAACYTCGGATGNCCAAAAAA	560-680-pb	DABERT (2008, 2010)
		LCO1490: GGTCAACAAATCATAAGATATTGG LCO2198: TAAACTTCAGGGTGACCAAAAAATCA		
Mites	18S	18S-1F (ATATTGGAGGGCAAGTCTGG) 18S-1R (TGGCATCGTTATGGTTAG)	500-pb	OTTO and WILSON, 2001

### 8.2.2 Morphological tools

The vouchers slide-mounted were identified up to genus, using the key proposed by Brennan and Goff (1977), and up to species based on the original descriptions of the species cited in the checklist from Brazil (Jacinavicius et al., 2018b) or comparing with type and material slides that were deposited at the United State National Museum (USNM), which are housed at the Systematic Entomology Laboratory, Beltsville, Maryland, USA (BARC-USDA-ARS).

## 8.3 RESULTS

A total of 317 chigger mites examined at the IBSP collection have been collected parasitizing nine cricetid rodents (*Akodon montensis*, *Akodon* sp., *Delomys sublineatus*, *Euryoryzomys russatus*, *Hylaeamys megacephalus*, *Necromys lasiurus*, *Nectomys squamipes*, *Oligoryzomys* sp. and *Thaptomys nigrita*), one echimyid rodent (*Trichomys fosteri*), and four marsupials (*Didelphis aurita*, *Gracilinanus agilis*, *Monodelphis americana*, *Monodelphis domestica* and *Thylamys macrurus*). After DNA extraction, mites were identified and generated 22 vouchers of *Arisocerus hertigi* Brennan, 1970, 14 of *Eutrombicula tinami* (Oudemans, 1910), 7 of *Kymocta* sp., 175 of *Quadraseta brasiliensis* Goff and Gettinger, 1989, 6 of *Quadraseta falconensis* Goff and Brennan, 1977, 5 of *Quadraseta flochi* (Brennan and Jones, 1960), 4 of *Quadraseta mackenziei* (Yunker and Brennan, 1964), 54 of *Quadraseta pazca* (Brennan and Jones, 1964), 11 of *Quadraseta trapezoides* (Brennan and Jones, 1964), 9 of *Quadraseta* sp., 1 of *Serratacarus* sp. and 9 of *Trombewingia bakeri* (Fonseca, 1955) (Appendix 1).

A total of 23 samples yielded 18S amplicons that generated DNA sequences (Appendix 1), which corresponded to a single haplotype for each species, from the following chiggers (GenBank accession number): one *A. hertigi* (MG817637), two *Kymocta* sp. (MG817642), four *Q. brasiliensis* (MG817643), two *Q. pazca* (MG817644), seven *Q. trapezoides* (MG817645), one *Quadraseta* sp. (KY934461) and three *T. bakeri* (MG817646). One of the amplified samples of *Q. pazca* did not generate a sequence of good quality and was not deposited in GenBank. Attempts to amplify fragments of the COI gene were unsuccessful (Appendix 1).

After a BLAST analysis, the consensus of these 18S sequences showed that the unique haplotype of each of the species *A. hertigi*, *Kymocta* sp., *Q. brasiliensis*, *Q. pazca*, *Q.*

*trapezoides*, *Quadrasetas* sp., and *T. bakeri* were 98.63% (431/437-pb), 99.51% (408/410-pb), 99.50% (397/399-pb), 99.75% (404/405-pb), 98.77% (400/405-pb), 98.39% (428/435-pb), and 99.50% (397/399-pb), respectively, identical to the corresponding sequence of *Eutrombicula splendens* (Ewing, 1913) (KY922159).

Once the endogenous control was tested, all 23 samples positive for 18S were submitted to the PCR assays targeting pathogens. A total of 13 samples yielded rickettsial *gltA* amplicons; eight of them were successfully sequenced. In addition, three of these samples also yielded rickettsial *ompA* amplicons. No sample yielded amplicons for the 16S (*Coxiella* sp.) and 18S (*Hepatozoon* sp.) genes, as shown on Appendix 1.

The rickettsial DNA sequences generated from the chiggers were submitted a BLAST analysis. Six *gltA* sequences showed to be 100% (350/350) identical to *Rickettsia* sp. strain Colombianensi" (MG970682). These sequences came from one larva of *A. hertigi*, three larvae of *Q. trapezoides* and two *T. bakeri*, regarding the haplotypes under accession numbers MG817637, MG817645 and MG817646, respectively. Other two *gltA* sequences were 99.43% (348/350-pb) and 100% (350/350-pb) identical to *Rickettsia parkeri* strain Atlantic Rainforest (MF536974). These sequences came from one larva of *A. hertigi* and one of *Q. trapezoides*, regarding the haplotypes under accession numbers MG817637 and MG817645, respectively. Besides that, for the two successfully sequenced-*ompA* samples, one was 100% (488/488) identical to *Rickettsia* sp. strain Colombianensi (MG970683) and the other was 98.37% (483/491) identical to *Rickettsia parkeri* strain Atlantic Rainforest (MF536975).

Two specimens of the species *A. hertigi* collected in the Zoo of São Paulo, São Paulo City, parasitizing *Oligoryzomys* sp., were found to be positive for *Rickettsia* sp. of two different strains, one *Rickettsia* sp. strain Colombianensi-like agent and another *Rickettsia parkeri* strain Atlantic rainforest-like agent. Three specimens of the species *Q. trapezoides* collected in Morro Grande, Cotia municipality, parasitizing *Nectomys squamipes*, were found to be positive for *Rickettsia* sp. strain Colombianensi-like agent, and another specimen on the same locality and host, were found to be positive for *Rickettsia parkeri* strain Atlantic rainforest-like agent. Besides that, two specimens of the species *T. bakeri* collected in Campos do Jordão municipality, parasitizing *Akodon montensis*, were found to be positive for *Rickettsia* sp. strain Colombianensi-like agent. In addition, we were able to amplify the corresponding rickettsial *ompA* gene for the strains detected in the species *A. hertigi*.

## 8.4 DISCUSSION

Regarding to the 18S gene of individually chiggers, the success rate of amplification for this gene was 13.63% (3/22) to *A. hertigi*, 28.57% (2/7) to *Kymocta* sp., 2.28% (4/175) to *Q. brasiliensis*, 5.55% (3/54) to *Q. pazca*, 63.64% (1/11) to *Q. trapezoides*, 11.11% (1/9) *Quadrasetata* sp., and 33.33% (3/9) to *T. bakeri*. To the species *E. tinami*, *Q. falconensis*, *Q. flocchi*, *Q. mackenziei* and *Serratacarus* sp. there was no success in obtaining 18S gene amplicons. For all species individually tested we were successful in amplifying 7.26% (23/317) of the samples, while Bassini-Silva et al (2018a) had 72.5% (29/40) successful in amplifying the same gene for the species *B. sinnamaryi* and Sang-Won Park et al. (2015) had 50% (38/76) for *Helenicula miyagawai* (Sasa, Kumada and Miura, 1951) and 87.5% (7/8) for *Leptotrombidium scutellare* (Nagayo, Miyagawa, Mitamura, Tamiya and Tenjin, 1921).

Among 73 species of chiggers from Brazil, 18S sequences were available for only the following three species: *B. sinnamaryi* (MG783391) (Bassini-Silva et al., 2018a), *Eutrombicula daemoni* Bassini-Silva and Jacinavicius, 2018 (MG707783, MG70778) (Bassini-Silva et al., 2018b), and *Q. brasiliensis* (MF113413, MF113412, KY934462, KY934463, KY934464) (Jacinavicus et al., 2018a). In the present study, we are providing new sequences of the 18S gene for the species *A. hertigi*, *Kymocta* sp., *Q. pazca*, *Q. trapezoides*, and *T. bakeri*. According to HILLIS and DIXON (1991) and CRUICKSHANK (2002), the 18S gene has a slow rate of evolution and it is suitable for phylogenies at the family and subfamily levels. Therefore, besides this gene is useful as an endogenous control of pathogen detection, the creation of a gene bank may contribute to the future studies of the relationships among mites in higher groupings.

The mitochondrial COI gene is known by the faster rate of sequence divergence than nuclear ribosomal genes (OTTO and WILSON, 1998). Kampen (2004), Moniuszko et al. (2015) and Kumlert et al. (2018) succeeded in amplifying this gene for some species of the chigger genera *Ascoshochengastia*, *Blankaartia*, *Hirsutiella*, *Leptotrombidium*, *Neotrombicula*, *Schoengastia*, *Schoutedenichia*, and *Walchia*, from Europe and Asia. However, in the present study, we tested the pairs of primers used by those authors and none of them amplified in our samples, similarly to previous studies in Brazil (Bassini-Silva et al. 2018a, Jacinavicius et al. 2018a).

Regarding to the detection of pathogens, in the present study we detected rickettsial DNA in eight specimens of chiggers from State of São Paulo for the first time. The only record of *Rickettsia* in chigger from Brazil was that of Bassini-Silva et al. (2018a) who

detected *Rickettsia felis*-like agent in *Blankaartia sinamaryi* parasitizing *Tachyphonus coronatus* in the state of Minas Gerais. The presence of *Rickettsia* spp. in mites collected on birds and mammals had already been previously recorded (VYSOTSKAYA and SLUGER, 1953; HASE et al., 1978; CHOI et al., 2007; TAKAHASHI et al., 2004; MITKOVA et al., 2015; HUANG et al. 2017; BASSINI-SILVA et al., 2018a). However, as stated by Bassini-Silva et al. (2018a), a deeper molecular characterization with DNA sequences of other genes are needed to confirm the *Rickettsia* species found in the chiggers. Notwithstanding, in the present study, the presence of *Coxiella* sp. and *Hepatozoon* sp. was not detected in the chiggers.

This is the first report of the detection *Rickettsia* spp. in chigger mites collected on rodents in Brazil. This group of mites has been neglected in the Neotropical region, mainly regarding to the pathogen infections, and more studies are needed to know if these mites are, in fact, involved in the epidemiological cycles of rickettsial diseases.

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**Appendix 1** - Results of the samples tested for the 18S\* and COI genes (endogenous control), *gltA* and *ompA* (*Rickettsia*), 16S (*Coxiella*) and 18S (*Hepatozoon*). A, amplified; NA, not amplified.

IBSP	Host	Locality	Stage	Species	18S*	COI	<i>gltA</i>	<i>OmpA</i>	16S	18S
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	A	NA	A	A	NA	NA
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	A	NA	A	NA	NA	NA
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	A	NA	A	A	NA	NA
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10519	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10520	<i>Oligoryzomys</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10521B	<i>Akodon</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10521B	<i>Akodon</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
10521B	<i>Akodon</i> sp.	São Paulo - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-
11239	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-

11239	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-	-
11239	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-	-
11239	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-	-
11239	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Arisocerus hertigi</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10535B	<i>Euryoryzomys russatus</i>	São Paulo - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
10541	<i>Didelphis aurita</i>	Peruíbe - SP	larva	<i>Eutrombicula tinami</i>	NA	NA	-	-	-	-	-
11129C	<i>Akodon</i> sp.	Cotia - SP	larva	<i>Kymocta</i> sp.	A	NA	A	NA	NA	NA	NA
11129C	<i>Akodon</i> sp.	Cotia - SP	larva	<i>Kymocta</i> sp.	A	NA	NA	-	NA	NA	NA
11998	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Kymocta</i> sp.	NA	NA	-	-	-	-	-

11999B	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Kymocta</i> sp.	NA	NA	-	-	-	-	-
11999B	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Kymocta</i> sp.	NA	NA	-	-	-	-	-
12001A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Kymocta</i> sp.	NA	NA	-	-	-	-	-
12001A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Kymocta</i> sp.	NA	NA	-	-	-	-	-
11130	<i>Didelphis aurita</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	A	NA	A	NA	NA	NA	NA
10530A	<i>Euryoryzomys russatus</i>	Peruíbe-SP	larva	<i>Quadraseta brasiliensis</i>	A	NA	NA	-	NA	NA	NA
12557A	<i>Akodon montensis</i>	Cotia - SP	ninfa	<i>Quadraseta brasiliensis</i>	A	NA	NA	-	NA	NA	NA
12557A	<i>Akodon montensis</i>	Cotia - SP	ninfa	<i>Quadraseta brasiliensis</i>	A	NA	NA	-	NA	NA	NA
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-

12557A	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
13070	<i>Hylaeamys megacephalus</i>	Brasília - DF	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
13070	<i>Hylaeamys megacephalus</i>	Brasília - DF	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12710B	<i>Akodon</i> sp.	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
11130	<i>Didelphis aurita</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
11110E	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
12706B	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
11110E	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10524A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10524A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10524A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10524A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10524A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10527A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10527A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10527A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-
10527A	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-







11996	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11997	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11997	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11997	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11997	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11997	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11305A	<i>Euryoryzomys russatus</i>	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11305A	<i>Euryoryzomys russatus</i>	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11305A	<i>Euryoryzomys russatus</i>	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11305A	<i>Euryoryzomys russatus</i>	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11305A	<i>Euryoryzomys russatus</i>	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11303C	<i>Oligoryzomys</i> sp.	Sete Barras - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11999A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11999A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-

11999A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11999A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11999A	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12000	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12001B	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12001B	<i>Delomys sublineatus</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12002	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12002	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12002	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12002	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12003	<i>Thaptomys nigrita</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12003	<i>Thaptomys nigrita</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-

12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12004	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12005	<i>Necromys lasiurus</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12006	<i>Didelphis aurita</i>	Itapevi - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12006	<i>Didelphis aurita</i>	Itapevi - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12006	<i>Didelphis aurita</i>	Itapevi - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-
12007	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta brasiliensis</i>	NA	NA	-	-	-	-	-

12007	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12007	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
12008	<i>Monodelphis americana</i>	Cotia - SP	larva	<i>Quadrasetas brasiliensis</i>	NA	NA	-	-	-	-	-
11700	<i>Thylamys macrurus</i>	Miranda - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
11724	<i>Gracilinanus agilis</i>	Terenos - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
11724	<i>Gracilinanus agilis</i>	Terenos - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
11724	<i>Gracilinanus agilis</i>	Terenos - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
11724	<i>Gracilinanus agilis</i>	Terenos - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
11724	<i>Gracilinanus agilis</i>	Terenos - MS	larva	<i>Quadrasetas falconensis</i>	NA	NA	-	-	-	-	-
13082	<i>Monodelphis domestica</i>	Cristino Castro - PI	larva	<i>Quadrasetas flochi</i>	NA	NA	-	-	-	-	-
13082	<i>Monodelphis domestica</i>	Cristino Castro - PI	larva	<i>Quadrasetas flochi</i>	NA	NA	-	-	-	-	-
13082	<i>Monodelphis domestica</i>	Cristino Castro - PI	larva	<i>Quadrasetas flochi</i>	NA	NA	-	-	-	-	-
13082	<i>Monodelphis domestica</i>	Cristino Castro - PI	larva	<i>Quadrasetas flochi</i>	NA	NA	-	-	-	-	-

13082	<i>Monodelphis domestica</i>	Cristino Castro - PI	larva	<i>Quadraseta flochi</i>	NA	NA	-	-	-	-
11720A	<i>Hylaeamys megacephalus</i>	Bonito - MS	larva	<i>Quadraseta mackenziei</i>	NA	NA	-	-	-	-
11720A	<i>Hylaeamys megacephalus</i>	Bonito - MS	larva	<i>Quadraseta mackenziei</i>	NA	NA	-	-	-	-
11720A	<i>Hylaeamys megacephalus</i>	Bonito - MS	larva	<i>Quadraseta mackenziei</i>	NA	NA	-	-	-	-
11720A	<i>Hylaeamys megacephalus</i>	Bonito - MS	larva	<i>Quadraseta mackenziei</i>	NA	NA	-	-	-	-
10530B	<i>Euryoryzomys russatus</i>	Peruíbe-SP	larva	<i>Quadraseta pazca</i>	A	NA	NA	-	NA	NA
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	A	NA	NA	-	NA	NA
11110G	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta pazca</i>	A	NA	NA	-	NA	NA
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12710A	<i>Akodon</i> sp.	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12706A	<i>Euryoryzomys russatus</i>	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
12710A	<i>Akodon</i> sp.	São Francisco do Sul - SC	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
10522	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-
10522	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadraseta pazca</i>	NA	NA	-	-	-	-



10529	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10529	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10529	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10530	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10530	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10530	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10530	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10530	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10531	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10531	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10531	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10531	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
10531	<i>Euryoryzomys russatus</i>	Peruíbe - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
11241A	<i>Akodon</i> sp.	Campos do Jordão - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
11241A	<i>Akodon</i> sp	Campos do Jordão - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
11241A	<i>Akodon</i> sp	Campos do Jordão - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
11241A	<i>Akodon</i> sp	Campos do Jordão - SP	larva	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
12557A	<i>Akodon montensis</i>	Cotia - SP	nymph	<i>Quadrasetapazca</i>	A	NA	NA	-	NA	NA	
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadrasetapazca</i>	NA	NA	-	-	-	-	-

12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadraseta</i> sp.	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadraseta</i> sp.	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadraseta</i> sp.	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadraseta</i> sp.	NA	NA	-	-	-	-	-
12805	<i>Akodon</i> sp	Cotia - SP	nymph	<i>Quadraseta</i> sp.	NA	NA	-	-	-	-	-
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	A	NA	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	NA	-	NA	NA	NA
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	A	NA	NA	-	NA	NA	NA
12557B	<i>Akodon montensis</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	NA	NA	-	-	-	-	-
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	NA	NA	-	-	-	-	-
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	NA	NA	-	-	-	-	-
11110C	<i>Nectomys squamipes</i>	Cotia - SP	larva	<i>Quadraseta trapezoides</i>	NA	NA	-	-	-	-	-
12489C	<i>Trichomys fosteri</i>	Nhecolândia - MS	larva	<i>Serratacarus</i> sp.	NA	NA	-	-	-	-	-
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	A	NA	A	NA	NA	NA	NA
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	A	NA	A	NA	NA	NA	NA
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	A	NA	A	NA	NA	NA	NA
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-	-

11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-
11226C	<i>Akodon montensis</i>	Campos do Jordão - SP	larva	<i>Trombewingia bakeri</i>	NA	NA	-	-	-	-

## 9. CONCLUSIONS

- This is the first description of *Q. brasiliensis* deutonymphs, developed from larvae maintained in laboratory conditions. Besides that, redescription of the larva was provided.
- In this study, the species *Arisocerus hertigi*, *Colicus spinosus*, *Eutrombicula alfreddugesi*, *Eutrombicula batatas*, *Eutrombicula goeldii*, *Eutrombicula tinami*, *Kymocta brasiliensis*, *Microtrombicula brachytrichia*, *Microtrombicula brennani*, *Microtrombicula rhipidomysi*, *Parasecia valida*, *Perissopalla tanydera*, *Quadraseta brasiliensis*, *Quadraseta pazca*, *Speleocola tamarina*, *Serratacarus dietzi* and *Trombewingia bakeri* are being recorded in new localities in Brazil.
- The presence of the chigger species *Boshkerria punctata*, *Eutrombicula spipi*, *Hooperella vesperuginis*, *Parascoshoengastia. aemulata*, *Quadraseta. azulae*, *Quadraseta falconensis*, *Quadraseta flochi*, *Quadraseta mackenziei*, *Quadraseta mirandae* and *Quadraseta trapezoides* are reported for the first time in Brazil.
- New associations of chiggers parasitizing mammals in Brazil have been provided, as following: Carnivora: *Nasua narica* and *Puma concolor*; Lagomorpha: *Sylvilagus brasiliensis*; Chiroptera: *Glossophaga soricina*, *Micronycteris (Neonycteris) pusilla*; Didelphimorpha: *Gracilinamus agilis*, *Monodelphis iheringi*, *Marmosa demerarae*, *Marmosa murina*; Rodentia: *Akodon cursor*, *Akodon montensis*, *Cerradomys subflavus*, *Cuniculus paca*, *Dasyprocta leporina*, *Euryoryzomys russatus*, *Kerodon rupestris*, *Oligoryzomys nigripes*, *Proechimys roberti*, *Rhipidomys macrurus*, *Sooretamys angouya*, *Thaptomys nigrita*, *Thricomys apereroides*, *Thrichomys fosteri* and *Thylamys macrurus*.
- Two new species were described, one for the genus *Parasecia* and another species for the genus *Pseudoschoengastia*.

- The genus *Arisocerus* was synonymized with the genus *Herpetacarus*. In addition, chiggers belonging to the genera *Boshkerria*, *Parascoshoengastia* and *Pseudoschoengastia* were recorded for the first time in Brazil.
- Finally, in this study, 73 species of chigger mites were recorded for the national territory.
- This is the first report of the detection of strains of *Rickettsia* sp. in chigger mites *A. hertigi*, *Q. trapezoides*, and *T. bakeri* collected on rodents in Brazil. In addition, sequences of 18S gene of the species *A. hertigi*, *Kymocta* sp., *Q. pazca*, *Q. trapezoides*, and *T. bakeri* were provided.