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A new genus and new species of macronyssid mite (Mesostigmata: Gamasina: Macronyssidae) from Brazilian caves including molecular data and key for genera occurring in Brazil

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Original research

ABSTRACT

A new genus and a new species of macronyssid mites is described based on material collected from caves in Minas Gerais state, Southeastern Brazil, and additional occurrences from caves in Rio de Janeiro (Southeast) and Pará (Eastern Amazon region). The new genus may be distinguished from other Macronyssidae genera by the combination of sternal shield with a distinct X-shaped area; sternal shield bearing three pairs of setae and two pairs of pores; sternal gland absent; anterior spur on coxa II lacking; adults with dorsal shield entire and j and J series complete; adults and protonymphs with setae J5 minute; males with strong-pores on dorsal shield and peritreme short extending from posterior margin of coxae II to the anterior margin of coxae IV. A key for genera occurring in Brazil is provided, as well sequences from the mitochondrial large subunit of ribosome (16S).

Keywords Acari; Dermanyssoidea; Ectoparasitic mites; Integrative taxonomy; key

Zoobank <http://zoobank.org/9C3C87C0-5371-4A8E-80B1-2F4E89632BDC>

Introduction

The family Macronyssidae Oudemans, 1936 includes mites that can be associated with a variety of vertebrates, such as birds, reptiles and mammals, including humans (Radovsky 2010; Orlova *et al.* 2017; Bassini-Silva *et al.* 2019). The life cycle of these mites comprises five stages of development: egg, larva, protonymph, deutonymph and adults (male or female) (Radovsky 1966; 1967). Of these, only protonymph and adults are hematophagous, other stages are inactive and can be found on the substrate or roost of hosts (Radovsky 2010).

Radovsky (2010) reviewed the Macronyssidae family, recognizing seven new genera for the family, from new species discovered by him or by splitting other genera, mainly *Ornithonyssus* Sambon, 1928, which included at least a third of the Macronyssid genera (Zumpt and Till 1953; Fonseca 1954; 1960; Radovsky 1966; 1969; Micherdzinski 1980; Shepherd and Narro 1983).

Currently, Macronyssidae comprises 34 valid genera, with occurrences worldwide. Of these, at least 23 genera occur in the Neotropical region associated with different host taxa: *Synasponyssus* Radovsky and Fuman 1969, *Parichoronyssus* Radovsky 1966, *Radfordiella* Fonseca 1948, *Chiroecetes* Herrin and Radovsky 1974, *Mitonyssoides* Yunker, Lukoschus and Giesen 1990, *Macronyssoides* Radovsky 1966, *Nycteronyssus* Saunders and Yunker 1973,

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Mitonyssus Yunker and Radovsky 1980, *Cryptonyssus* Radovsky 1966, *Chiroptonyssus* Augustson 1945, and *Macronyssus* Kolenati 1858 are associated exclusively with bats; *Steatonyssus* Kolenati 1858 with bats and other mammals; *Acanthonyssus* Yunker and Radovsky 1966, *Argitis* Yunker and Saunders 1973, *Lepidodorsum* Saunders and Yunker 1975, *Lepronyssoides* Fonseca 1941 and *Lagidonyssus* Radovsky 2010 with rodents, despite *Lepronyssoides* can be found associated with other mammals; *Pellonyssus* Clark and Yunker 1956 with birds; *Ornithonyssus* Sambon 1928 with birds and mammals, including humans; *Draconyssus* Yunker and Radovsky 1966, *Ophionyssus* Megnin 1884, *Endophionyssus* Radovsky 2010 on lizards and snakes; and, finally, *Thigmonyssus* Radovsky 2010 found associated with anteaters.

Thirteen macronyssid genera are recorded from Brazil thus far: *Chiroptonyssus*, *Lepronyssoides*, *Macronyssoides*, *Macronyssus*, *Mitonyssoides*, *Mitonyssus*, *Ophionyssus*, *Ornithonyssus*, *Parichoronyssus*, *Pellonyssus*, *Radfordiella*, *Steatonyssus* and *Thigmonyssus* (Bassini-Silva *et al.* 2020). They comprise twenty-seven species of mites occurring in localities in the Southeast (Minas Gerais, Rio de Janeiro and São Paulo), Middle-west (Mato Grosso, Mato Grosso do Sul and Goiás), Northeast (Alagoas, Bahia, Ceará, Maranhão, Paraíba, Pernambuco, Piauí and Rio Grande do Norte) and South (Paraná and Rio Grande do Sul) (Fonseca 1948; Fonseca 1954; Radovsky 1967; Whitaker Jr and Mumford 1977; Azevedo *et al.* 2002; Almeida *et al.* 2011; Nieri-Bastos *et al.* 2011; Moras *et al.* 2013; Silva *et al.* 2017; Bassini-Silva *et al.* 2019; 2020; Oliveira *et al.* 2020).

Herewith, we aim at describing a new genus and new species based on adults and protonymphs found in caves located in the states of Minas Gerais, Rio de Janeiro (Southeastern) and Pará (Eastern Amazon region).

Material and methods

Specimens were collected during surveys for the subterranean fauna, in iron ore and limestone caves in Minas Gerais and Rio de Janeiro states, Southeastern Brazil and from caves in Pará state, North Brazil (Figure 1A). Mites were found freely dwelling on the cave floor, from entrance, penumbra and aphotic zone, during the dry and rainy seasons (Table 1; Figure 1B).

Specimens were mounted as permanent microscope slides using Hoyer's medium. Measurements and illustrations were made using the Leica DM 750 optical microscope with an ICC50 W digital camera attached. All measurements are given in micrometers (µm). Illustrations were prepared using the live image overlay technology adapted from Sidorchuk and Vorontsov (2014). Map was produced with QGIS 3.18.1 program and figures were prepared with Krita v.4.4.2 program. Terminology of dorsal and ventral chaetotaxy and the nomenclature of the dorsum based on Radovsky (2010). The terminology of sternal pores was reported according to Evans and Till (1979).

Holotype, paratypes and additional individuals (Supplementary material 1) are deposited at the Acarological Collection, Centro de Coleções Taxonômicas, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Belo Horizonte City. Collection acronym: UFMG AC.

Molecular procedures

DNA extraction — Specimens were preserved in 95–100 % ethanol and stored at –20 °C. Genomic DNA was extracted from single specimens using a QIAamp DNA Micro kit (Qiagen) following the manufacturer's protocol, except by using two steps of the final elution, leading to a final volume of 50 µl.

16S rRNA gene amplification — A fragment of 16S rRNA was chosen due its availability for other Macronyssidae, since was employed in previous studies on the Macronyssid genus *Ornithonyssus*. The amplification of mitochondrial large ribosomal subunit (16S) fragment of ~ 410 bp was conducted using the primers proposed by Mangold *et al.* (1998): 16S + 1 (5'-CTG CTC AAT GAT TTT TTA AAT TGC TGT GG-3'); 16S–1 (5' -CCG GTC TGA ACT CAG ATC AAG T-3'). Amplifications were performed in 20 µl of final volume, with Platinum

Figure 1 A – Sample distribution map with distribution across Brazilian territory and details of the distribution in the state of Pará and Minas Gerais, respectively; B – Graphic detail of the cave zone and period of collection of 39 specimens, of a total of 188 samples examined; C – Maximum Likelihood Tree inferred from sequences of mitochondrial 16S from Macronyssidae deposited in GenBank, in addition to sequences obtained from individuals of *Chiasmanyssus cavernicola* n. sp. Values above branches are Bootstrap support and below Posterior Probabilities (both in %). Clades absent in the Bayesian analyses are indicated by “–”.

Table 1 Detailed sampling data of specimens with zoning information in the cave, season or collection period. Abbreviations: Protonymph (PN) female (F), male (M), Minas Gerais state (MG).

AC-UFGM	Instar/Sex	Cave	Zone	Season	Date	Latitude/Longitude	Municipality	State
171097	F	CAI-03	Entrance	-	27-29/II/01-09/III/2012	18°56'10.6"S 43°24'45.5"W	Conceição do Mato Dentro	MG
174528	F	CCB-0052	Penumbra	Humid	22-24/I/2018	20°09'58.9"S 43°22'59.7"W	Mariana	MG
174529	F	CCB-0052	Penumbra	Humid	22-24/I/2018	20°09'58.9"S 43°22'59.7"W	Mariana	MG
174550	F	CCB-0052	Penumbra	Dry	11-13/IX/2018	20°09'58.9"S 43°22'59.7"W	Mariana	MG
174537	F	CCB-0063	Penumbra	Dry	11-13/IX/2017	20°09'58.1"S 43°23'16.8"W	Mariana	MG
174554	F	CCB-0063	Penumbra	Dry	11-13/IX/2018	20°09'58.1"S 43°23'16.8"W	Mariana	MG
180365	F	CSS-0001	Penumbra	Humid	30/IV a 05/V/2018	18°55'02.1"S 43°25'43.7"W	Conceição do Mato Dentro	MG
180368	M	CSS-0001	Penumbra	Humid	30/IV a 05/V/2018	18°55'02.1"S 43°25'43.7"W	Conceição do Mato Dentro	MG
180371	F	CSS-0001	Penumbra	Humid	30/IV a 05/V/2018	18°55'02.1"S 43°25'43.7"W	Conceição do Mato Dentro	MG
180376	F	CSS-0001	Penumbra	Humid	30/IV a 05/V/2018	18°55'02.1"S 43°25'43.7"W	Conceição do Mato Dentro	MG
180348	F	CSS-0005	Penumbra	Humid	30/IV a 05/V/2018	18°55'03.8"S 43°25'42.7"W	Conceição do Mato Dentro	MG
180364	F	CSS-0005	Penumbra	Humid	30/IV a 05/V/2018	18°55'03.8"S 43°25'42.7"W	Conceição do Mato Dentro	MG
180339	F	CSS-0006	Aphotic	Humid	30/IV a 05/V/2018	18°55'04.3"S 43°25'43.6"W	Conceição do Mato Dentro	MG
180340	M	CSS-0007	Penumbra	Humid	30/IV a 05/V/2018	18°55'08.0"S 43°25'13.6"W	Conceição do Mato Dentro	MG
180341	M	CSS-0007	Penumbra	Humid	30/IV a 05/V/2018	18°55'08.0"S 43°25'13.6"W	Conceição do Mato Dentro	MG
180361	F	CSS-0007	Penumbra	Humid	30/IV a 05/V/2018	18°55'08.0"S 43°25'13.6"W	Conceição do Mato Dentro	MG
180367	F	CSS-0084	Penumbra	Humid	30/IV a 05/V/2018	18°55'00.3"S 43°25'48.0"W	Conceição do Mato Dentro	MG
170372	F	CSS-0111	Entrance	-	31/V-12/VI/2016	18°23'48.7"S 43°24'49.3"W	Conceição do Mato Dentro	MG
171374	F	HOLC-0090/97	Aphotic	Humid	21/XI-02/XII/2016	19°36'10.5"S 44°00'43.1"W	Pedro Leopoldo	MG
171375	F	HOLC-0090/97	Aphotic	Humid	21/XI-02/XII/2016	19°36'10.5"S 44°00'43.1"W	Pedro Leopoldo	MG
180121	F	MCFC-0009	Aphotic	Dry	26/VI-06/VII/2018	19°19'52.2"S 43°18'41.8"W	Itambé do Mato Dentro	MG
180093	PN	MCFC-0012	Aphotic	Dry	26/VI-06/VII/2018	19°20'35.0"S 43°18'28.8"W	Itambé do Mato Dentro	MG
180094	F	MCFC-0012	Aphotic	Dry	26/VI-06/VII/2018	19°20'35.0"S 43°18'28.8"W	Itambé do Mato Dentro	MG
180106	F	MCFC-0013	Entrance	Dry	26/VI-06/VII/2018	19°20'38.8"S 43°18'16.8"W	Itambé do Mato Dentro	MG
180419	F	MGB-0061	Penumbra	Dry	I/VIII/2018	20°26'37.0"S 43°45'55.1"W	Ouro Preto	MG
180473	F	MGB-0063	Penumbra	Dry	03-06/IX/2018	20°26'37.2"S 43°45'55.3"W	Ouro Preto	MG
180483	F	MGB-0063	Penumbra	Dry	03-06/IX/2018	20°26'37.2"S 43°45'55.3"W	Ouro Preto	MG
180489	F	MGB-0063	Aphotic	Dry	03-06/IX/2018	20°26'37.2"S 43°45'55.3"W	Ouro Preto	MG
180067	F	MS-0004	Aphotic	Humid	26-28/III/2018	20°11'01.7"S 43°58'19.8"W	Nova Lima	MG
180073	F	MS-0004	Aphotic	Dry	28-29/IX/2017	20°11'01.7"S 43°58'19.8"W	Nova Lima	MG
180074	PN	MS-0004	Aphotic	Dry	28-29/IX/2017	20°11'01.7"S 43°58'19.8"W	Nova Lima	MG
180076	F	MS-0004	Penumbra	Dry	28-29/IX/2017	20°11'01.7"S 43°58'19.8"W	Nova Lima	MG
180077	F	MS-0004	Penumbra	Dry	28-29/IX/2017	20°11'01.7"S 43°58'19.8"W	Nova Lima	MG
180065	F	MS-0025	Entrance	Humid	26-28/III/2018	20°14'52.7"S 43°57'59.6"W	Itabirito	MG
180068	F	MS-0025	Entrance	Humid	26-28/III/2018	20°14'52.7"S 43°57'59.6"W	Itabirito	MG
180052	F	MS-0030	Penumbra	Humid	26-28/III/2018	20°12'07.8"S 43°58'03.7"W	Nova Lima	MG
180053	F	MS-0030	Aphotic	Humid	26-28/III/2018	20°12'07.8"S 43°58'03.7"W	Nova Lima	MG
180049	PN	MS-0031	Aphotic	Humid	26-28/III/2018	20°12'08.7"S 43°58'03.4"W	Nova Lima	MG
180050	F	MS-0031	Aphotic	Humid	26-28/III/2018	20°12'08.7"S 43°58'03.4"W	Nova Lima	MG

Taq DNA Polymerase (Invitrogen) in a Mastercycler nexus (Eppendorf) thermocycler. Master mix for initial PCR contained 2.0 µl of PCR buffer (1X), 1.4 µl MgCl₂ (50 mM), 1.4 µL of dNTP (10 mM each), 0.8 µl of each oligonucleotide primer. Amplifications used 1–3 µl of genomic DNA and included an initial denaturing step of 2 min at 94 °C, followed by 35 cycles, comprising: denaturing at 94 °C for 30s, annealing for 35s, and extension at 72 °C for 45 s. The annealing temperature for the first seven cycles increased in 0.3 °C every cycle, ranging from 47 °C to 48.8 °C, remaining 28 cycles used an annealing temperature of 50 °C. Final extension of 5 min, at 72 °C.

Sequencing and editing chromatogram — PCR products found positive in 1% agarose gel electrophoresis were purified using the Ampure® (Agencourt) kit and sequenced using a 3730 DNA Analyzer. Sequencing primers as for PCRs. Chromatograms were resolved in ChromasPro 1.41 (Technelysium Pty Ltd).

Sampling, alignment and 16S secondary structure — Newly obtained sequences and those downloaded from GenBank are reported in Table 2, along voucher material collection numbers. This fragment was chosen because it led to the largest sample of terminals identified to species and with vouchers data. It is beyond the objectives of this article perform a

throughout phylogenetic analyses of Macronyssidae, something that would require more markers and terminals. The aim of including this fragment is just to verify if it supports the inclusion of the new genus in the relatively well sampled *Ornithonyssus*. Preliminary automatic alignment was performed using MUSCLE (Edgar 2004) and then manually aligned with aid of sequence editor BioEdit 7.2.1 (Hall 1999) considering the 16S secondary structure as reported for other arthropods (Smith and Bond 2003; Gillespie *et al.* 2006). A full annotated alignment is provided in the supplementary material. Therein, matching parentheses and dots were used to indicate stem and loop regions, respectively; structural helix numbering is given after Gillespie *et al.* (2006). Length variable regions inferred to be ambiguously aligned were classified into regions of expansion and contraction (REC), non-pairing regions of ambiguous alignment (RAA) and excluded from downstream phylogenetic analyses. Access to the Brazilian genetic heritage was registered in SisGen under register number A68A31E.

Molecular Phylogenetic Analysis — Model for sequence evolution was chosen using the AICc criterion implemented in jModeltest (Darriba *et al.* 2012). Maximum Likelihood inference was run in PhyML (Guindon *et al.* 2010) online at <http://www.atgc-montpellier.fr/phyml/> (accessed January 18nd 2021) and Bayesian analyses in MrBayes 3.2.7 (Ronquist *et al.* 2012).

Table 2 GenBank accession numbers and vouchers for 16S gene. The sequence of the Rhinonyssid *Ptilonyssus chloris* (Rhinonyssidae) was used as outgroup in some analyses. * In Radovsky (2010) the current taxonomic position of this species is placed in the genus *Lepronyssoides*.

Terminal	Voucher	GenBank	Terminal	Voucher	GenBank
Rhinonyssidae			<i>Ornithonyssus bursa</i>	LEcEN SIA Ornis31	MK131067
<i>P. chloris</i> (Fain 1962)		NC_045208		LEcEN SIA Ornis14	MK131059
Macronyssidae				LEcEN SIA Ornis51	MK131071
<i>Pellonyssus cf. reedi</i>	LEcEN: SIA:Ornis70	MK131079		LEcEN SIA Ornis32	MK131068
	IBSP10372	HM059863		LEcEN SIA Ornis109	MK131086
	IBSP10371	HM059862		LEcEN SIA Ornis102	MK131085
	IBSP10370	HM059861		LEcEN SIA Ornis101	MK131084
	IBSP10369	HM059860		LEcEN SIA Ornis99	MK131083
	IBSP10368	HM059859		LEcEN SIA Ornis98	MK131082
	IBSP10367	HM059858		LEcEN SIA Ornis97	MK131081
	IBSP10366	HM059857		LEcEN SIA Ornis72	MK131080
<i>Ornithonyssus brasiliensis</i> (Fonseca 1939)	IBSP10365	HM059856		LEcEN SIA Ornis69	MK131078
	IBSP10364	HM059855		LEcEN SIA Ornis67	MK131077
	IBSP10363	HM059854		LEcEN SIA Ornis65	MK131076
	IBSP10362	HM059853		LEcEN SIA Ornis55	MK131074
	IBSP10361	HM059852	<i>Ornithonyssus sylviarum</i>	LEcEN SIA Ornis54	MK131073
	IBSP10360	HM059851		LEcEN SIA Ornis52	MK131072
	IBSP10359	HM059850		LEcEN SIA Ornis48	MK131070
	IBSP10358	HM059849		LEcEN SIA Ornis39	MK131069
	IBSP10357	HM059848		LEcEN SIA Ornis27	MK131066
<i>Ornithonyssus vitzthumi</i> (Fonseca 1941)	IBSP8546	HM059847		LEcEN SIA Ornis26	MK131065
	IBSP10356	HM059846		LEcEN SIA Ornis24	MK131064
	IBSP10355	HM059845		LEcEN SIA Ornis23	MK131063
	IBSP10354	HM059844		LEcEN SIA Ornis22	MK131062
	IBSP10353	HM059843		LEcEN SIA Ornis21	MK131061
	IBSP10352	HM059842		LEcEN SIA Ornis15	MK131060
	IBSP10351	HM059841		LEcEN SIA Ornis4	MK131057
<i>Ornithonyssus wernecki</i>	IBSP10348	HM059837		LEcEN SIA Ornis2	MK131056
	IBSP10347	HM059836	<i>Ornithonyssus</i> sp.	LEcEN SIA Ornis91	MK131087
	IBSP10346	HM059835	<i>Chiasmanyssus cavernicola</i> n. gen. n. sp.	UFMG AC 200347	MW522521
<i>Lepronyssoides pereirai</i> *	IBSP8770	HM059834		UFMG AC 200348	MW522522
<i>Ornithonyssus bacoti</i>	IBSP10350	HM059840		UFMG AC 200349	MW522523
	IBSP10349	HM059839		UFMG AC 200350	MW522524
<i>Ornithonyssus monteiroi</i>	IBSP10388	HM059838		UFMG AC 200380	MW522525
				UFMG AC 200381	MW522526
				UFMG AC 200382	MW522527
				UFMG AC 200383	MW522528
				UFMG AC 200384	MW522529
				UFMG AC 200385	MW522530
				UFMG AC 200386	MW522531
				UFMG AC 200387	MW522532

Inter and intraspecific distances were calculated in MEGA7 (Kumar *et al.* 2016).

Result

DNA sequencing and analyses

The final alignment counted with 288 positions after having their 3' and 5' extremities trimmed, and regions of ambiguous alignment excluded. Among them, 186 positions were conserved across all macronyssid species and 78 parsimony informative. Whereas intraspecific mean p-distances among sequences varied from 0.0005 in *O. sylvarum* (Canestrini and Fanzago 1877) (n= 24) to 0.0094 in *O. wernecki* (Fonseca 1935a) (n= 3), among *Ornithonyssus* and *Lepronyssoides* species, interspecific p-distances varied from 0.0462 (*O. bacoti* (Hirst 1913) x *O. monteroi* (Fonseca 1941) and 0.1435 (*O. bursa* (Berlese 1888) x *O. wernecki* and *L. pereirai* (Fonseca 1935a). Pairwise mean interspecific p-distances are summarized in Table 3. The results suggest the 16S fragment sequenced here is a useful barcoding marker for molecular identification.

The model HKY+G was chosen using jModeltest and used in Maximum likelihood and Bayesian analyses. They agree to each other and similar to Nieri-Bastos *et al.* (2011) recovers *L. pererai* and *O. wernecki* in a basal position relative to remaining *Ornithonyssus* species (Figure 1C). *Chiasmanyssus cavernicola* **n. gen. n. sp.** branches basal in the topology obtained, being recovered in an isolated position relative to *Ornithonyssus* and *Lepronyssoides* species from which 16S sequences are available.

Taxonomy

Family Macronyssidae Oudemans, 1936

Diagnosis according to Radovsky (2010).

Chiasmanyssus n. gen.

Zoobank: 1C2CF818-9683-4DB2-BE34-DAD69DC435F5

(Figures 3–10)

Type species: *Chiasmanyssus cavernicola*, new species.

Diagnosis — Female large, its sternal shield square, distinctly sclerotized, with a smoother area on central sternal shield X-shaped; sternal shield bearing three pairs of setae and two pairs of pores; without sternal glands; anterior median protusion in epigynal shield extends under the sternal shield; anterior spur on coxa II lacking. Adults with dorsal shield entire, reticulated

Table 3 Pairwise mean interspecific p-distances values between sequences of 16S from species of macronyssid mites deposited in GenBank and newly sequenced from the new genus. Values with cells shadowed in gray are intra-specific mean distances in case of more than one sequence available for that species.

	1	2	3	4	5	6	7	8	9	10	11
1. <i>Pellonyssus</i>	--										
2. <i>O. brasiliensis</i>	0.1828	0.0013									
3. <i>O. vitzhumi</i>	0.1816	0.0602	0.0078								
4. <i>O. wernecki</i>	0.1799	0.1274	0.1094	0.0094							
5. <i>L. pereirai</i>	0.1825	0.1239	0.1156	0.1115	--						
6. <i>O. bacoti</i>	0.1707	0.0609	0.0561	0.1162	0.0943	0.0036					
7. <i>O. monteroi</i>	0.1944	0.0605	0.0732	0.1256	0.1021	0.0462	--				
8. <i>O. bursa</i>	0.1815	0.0667	0.0839	0.1435	0.1435	0.0738	0.0731	0.0018			
9. <i>O. sylvarum</i>	0.1825	0.0545	0.0726	0.1254	0.1297	0.0628	0.0632	0.0476	0.0005		
10. <i>Ornithonyssus</i> sp.	0.1905	0.0700	0.0550	0.1176	0.1163	0.0686	0.0504	0.0921	0.0778	--	
11. <i>C. cavernicola</i> n. gen. n. sp.	0.1620	0.1479	0.1432	0.1567	0.1543	0.1381	0.1437	0.1569	0.1554	0.1388	0.0017

and with j and J series complete. Adults and protonymphs with setae J5 minute; all setae on idiosoma, legs and gnathosoma slender and smooth, except palp with disto-ventral seta on genu spatulate. Females and protonymphs with chelae simple and slender, without spines or other processes. Male with four pairs of conspicuous pores near setae j3, z4, J3 and one laterally between J4 and J5 setae; peritrema short, extending from posterior margin of coxae II to the anterior margin of coxae IV and peritrematic shield with nodule fused to the middle of the shield.

Etymology — The generic name comes from Greek word “*chiasma*” referring to the distinct X-shaped area on sternal shield and “*nyssus*”, to prick, referring to the parasitic life.

***Chiasmanyssus cavernicola* n. gen. n. sp.**

Zoobank: [A34F4774-19C6-41E7-AFCB-023910A345A0](https://zoobank.org/A34F4774-19C6-41E7-AFCB-023910A345A0)

Diagnosis — *Female*: Elongated dorsal shield, holotrichous, bearing 27 pairs of setae, all setae long and subequal, except j2, z2, s1–s2 small (and J5 minute). Unarmed dorsum with approximately 84 pairs of long setae and ventral opisthosoma with approximately 48 pairs of setae; peritrema long, extending from level of anterior margin of coxae II to the middle of coxae IV; palpal trochanter with a small blade-like ventral process. *Male*: anterior margin of the dorsal shield not discernible from the soft cuticle; with neotrichous condition on opisthonotal region; a pair of additional minute setae, laterally to J4 and J5, on neotrichous area of opisthonotal region. Four pairs of caudal setae on unarmed dorsum and four pairs of setae on unarmed venter. *Protonymph*: Subrectangular podonotal shield with lateral borders biconcave at level of z2 and z4, posterior region almost square; with j2 very close to j1. Trapezoidal pygidial shield with J4 and Z4 small than others (except J5 minute). Unarmed dorsum with 12 pairs of long setae. Unarmed venter with four pairs of median setae between ventral shields and a pair of caudal setae, without setae flanking the anus.

Description. Female — Measurements summarized in table 4.
(Figures 2–4)

Idiosoma — (Figures 2A–B). Oval, moderately sclerotized and striated integument; all setae smooth. *Dorsum* (Figure 2A). Dorsal shield entire, elongated, longer than wide, and narrow, even at its widest, distinctly tapered in the middle of opisthonotal region; reticulated sculpture on all its area; holotrichous condition with 27 pairs of setae, 16 on podonotum and 11 on opisthonotum; j and J series complete with j1, j2 and j3 on dorsal shield; all setae long and subequal except j2, z2, s1–s2 small and J5 as microsetae (Figure 3A); one pair of lyrifissures near on margin lateral to setae j1 and four pair of pores on margin lateral to setae j3 and J4 and near s4 and z6. Unarmed dorsum with approximately 84 pairs of long setae. *Venter* (Figure 2B). Tritosternal base not expanded; Tritosternum bipartite with long and fimbriate laciniae (Figure 3B). Presternal area is weakly sculptured. Sternal shield nearly square, anterior margin with a small medial prominence; strongly sclerotized and punctate, except for a smoother X-shaped portion on central sternal shield (Figure 3C); anterior and posterior corners absent; three pairs of sternal setae relatively long, st1 and st2 on the sclerotized part and st3 inserted on X-shaped region on shield; two pairs of sternal pores. Metasternal setae (st4) on unarmed integument with third pair of pores accompanying metasternal setae. Epigynal shield expanded anteriorly, median projection extends under the sternal shield, no anterior scale-like markings; posterior margin pointed (Figure 3C); one pair of genital setae, approximately at the middle of epigynial plate. Anal plate pyriform; long and narrow cribrum; one pair of adanal setae (pan) and one postanal seta (pon) (Figure 3D). Endopodal plates small and triangular between level of coxae II and III. Opisthosoma with approximately 48 pairs of setae ventrally, slender, except caudal setae, being thicker, similar to the setae on unarmed dorsum. Peritreme long, extending from anterior margin of coxae II to the middle of coxae IV (Figure 2B). Peritrematic shield free from dorsal shield but interrupted by two “nodules” on the anterior dorsum between coxae I and II and in the middle ventral between coxae II and III (Figure 2A–B). Peritrematic shield posterior to the stigma fused with the parapodal shield around the posterior margin of coxa IV (Figure 2B).

Table 4 Metric data of females, males and protonymphs specimens from new genus.

Characters	FEMALE				MALE			PROTONYMPH		
	Holotype	n	range	mean	n	range	mean	n	range	mean
idiosoma L	792	10	726-847	782	2	520-693	606	2	400-424	412
idiosoma W.	432	10	377-507	451	2	294-384	339	2	264-288	276
dorsal shield L.	624	10	569-658	625	2	534-605	570	-	-	-
anterior dorsal plate W.	-	-	-	-	2	216-233	225	-	-	-
middle dorsal plate W.	-	-	-	-	2	313-319	316	-	-	-
posterior dorsal plate W.	-	-	-	-	2	79-93	86	-	-	-
dorsal shield W. at j3	199	10	191-218	202	-	-	-	-	-	-
dorsal shield W. at j6	202	10	173-225	202	-	-	-	-	-	-
dorsal shield W. at J5	41	10	33-42	38	-	-	-	-	-	-
podonotal shield L.	-	-	-	-	-	-	-	2	204-211	207
podonotal shield W. at z2	-	-	-	-	-	-	-	2	132-139	136
podonotal shield W. at j5	-	-	-	-	-	-	-	2	176-184	180
podonotal shield W. at j6	-	-	-	-	-	-	-	2	132-134	133
anterior pygidial shield L.	-	-	-	-	-	-	-	2	71-73	72
pygidial shield W. at Z3	-	-	-	-	-	-	-	2	107-112	109
pygidial shield W. at S5	-	-	-	-	-	-	-	2	86-89	88
pygidial shield W. at Z5	-	-	-	-	-	-	-	2	49-60	54
j1	51	10	40-61	52	2	27-33	30	2	33-38	35
j2	22	10	22-32	27	2	18-19	19	2	24-29	27
j3	54	9	43-59	52	2	30-37	34	2	40-42	41
j4	65	9	64-76	68	1	57	-	2	42-57	49
j5	61	7	60-78	66	2	31-47	39	2	51-64	58
j6	85	9	69-94	83	1	60	-	2	80-80	80
z2	27	10	24-32	28	2	23-24	24	2	20-23	21
z4	54	8	45-59	52	1	39	-	2	60-63	62
z5	83	9	64-86	78	1	56	-	2	64-64	64
z6	75	10	68-86	74	1	55	-	-	-	-
s1	28	10	23-34	28	-	-	-	-	-	-
s2	32	10	24-37	31	2	45-49	47	-	-	-
s3	60	10	58-76	67	2	52-94	73	-	-	-
s4	63	10	63-83	73	1	55	-	2	69-74	72
s5	70	8	58-72	66	2	52-52	52	-	-	-
s6	70	9	52-74	68	2	45-60	53	2	91-92	91
J1	81	9	65-82	76	1	62	-	2	81-89	85
J2	73	8	65-79	74	1	56	-	2	87-97	92
J3	63	8	50-70	60	1	57	-	2	96-96	96
J4	55	6	42-61	53	1	43	-	2	18-20	19
J5	-	-	-	-	1	10	-	-	-	-
Z1	71	10	59-76	68	-	-	-	-	-	-
Z2	61	7	57-71	65	-	-	-	-	-	-
Z3	56	7	46-57	54	-	-	-	2	92-96	94
Z4	47	6	33-48	41	-	-	-	2	10-10	10
Z5	73	9	62-84	72	1	60	-	2	102-106	104
S5	56	8	54-64	59	-	-	-	2	88-92	90
peritreme to stigma	238	10	213-254	234	2	104-106	105	2	62-62	62
sternal plate L. at st.3	124	10	112-124	118	-	-	-	2	160-161	161
sternal plate W.at st.1	133	10	129-146	137	-	-	-	2	65-70	67
sternal plate W. at st.2	121	10	117-129	122	-	-	-	2	96-97	96
sternal plate W.at st.3	126	10	122-132	127	-	-	-	2	69-70	70
holoventral shield L.	-	-	-	-	1	468	-	-	-	-
holoventral shield W. at st.2	-	-	-	-	2	87-93	90	-	-	-
holoventral shield W. at gen.	-	-	-	-	2	44-45	44	-	-	-
holoventral shield W. at pan.	-	-	-	-	1	52	-	-	-	-

Table 4 Continued

Characters	FEMALE				MALE			PROTONYMPH		
	Holotype	n	range	mean	n	range	mean	n	range	mean
st.1	52	8	50-63	55	2	31-36	34	2	42-44	43
st.2	52	7	46-56	52	2	27-32	30	2	44-45	44
st.3	59	7	50-83	64	2	35-41	38	2	50-50	50
st.4	60	7	50-64	57	2	27-41	34	2	33-33	33
epigynial plate W.	66	10	65-70	67	-	-	-	-	-	-
epigynial plate L.	292	10	289-321	306	-	-	-	-	-	-
genital setae L.	42	7	35-44	40	-	-	-	-	-	-
anal shield L.	137	10	109-137	125	-	-	-	2	68-71	69
anal shield W.	59	10	57-80	68	-	-	-	2	43-44	43
pan. setae	45	7	38-50	45	1	32	-	2	50-60	55
pon. setae	51	9	39-59	51	2	33-37	35	2	54-56	55
tarsus I L.	312	10	259-324	308	2	212-220	216	2	203-213	208
tibia I	150	10	130-154	144	2	101-104	103	2	84-88	86
Genu I	127	10	110-139	125	2	89-95	92	2	71-80	75
femur I	183	10	170-202	185	2	104-134	119	2	105-107	106
trocanter I	73	10	61-74	69	2	41-45	43	2	48-49	49
coxa I	95	10	87-97	93	2	64-74	69	2	62-70	66
leg I L.	940	10	855-958	914	2	615-667	641	2	572-607	590
tarsu II	223	10	186-239	218	2	128-133	130	2	143-148	145
tibia II	111	10	93-112	104	2	72-77	75	2	68-69	68
genu II	110	10	81-113	97	2	60-61	61	2	66-69	68
femur II	150	10	124-154	143	2	93-101	97	2	85-93	89
trocanter II	69	10	46-69	60	2	48-50	49	2	42-44	43
coxa II	61	10	45-74	61	2	39-53	46	2	30-33	31
leg II L.	724	10	615-738	683	2	442-473	458	2	440-449	445
tarsu III	219	10	182-233	217	2	111-137	124	2	138-139	139
tibia III	86	10	84-107	96	2	68-70	69	2	62-64	63
genu III	78	10	78-92	84	2	61-71	66	2	58-60	59
femur III	116	10	110-137	123	2	74-93	84	2	82-84	83
trocanter III	64	10	58-74	65	2	48-52	50	2	45-50	47
coxa III	68	10	58-77	67	2	44-50	47	2	34-41	38
leg III L.	630	10	598-685	652	2	416-463	439	2	425-433	429
tarsu IV	307	10	260-315	300	2	176-188	182	2	200-201	200
tibia IV	140	10	132-148	140	2	93-99	96	2	87-90	89
genu IV	119	10	113-137	124	2	76-92	84	2	81-82	81
femur IV	186	10	168-208	188	2	121-125	123	2	106-123	115
trocanter IV	80	10	80-107	89	2	63-77	70	2	70-73	71
coxa IV	66	10	58-81	67	2	52-53	53	2	38-46	42
leg IV L.	899	10	858-968	908	2	582-634	608	2	586-611	598
tritosternum L.	214	9	197-225	211	1	113	-	2	104-132	118
hyp.1	42	10	40-49	44	2	29-29	29	2	25-30	27
hyp.2	35	10	30-44	37	2	22-23	23	2	19-25	22
hyp.3	43	9	34-50	43	2	31-32	32	2	31-36	33
gnathosoma setae (pc)	44	10	44-56	49	2	28-29	28	2	26-34	30
chelicera L. with chela	223	10	213-231	224	2	127-129	128	2	152-159	156
spermatheca	-	-	-	-	2	37-40	38	-	-	-
palps	258	10	235-266	252	2	200-225	212	2	227-244	236

Figure 2 *Chiasmanyssus cavernicola* n. sp. female. A – Idiosomatic dorsum; B – Idiosomatic ventral. Scale Bars: A & B 20 μ m.

Legs — Legs long and slender; legs I and IV longer than others; leg setae smooth. Coxae II and III may have posterior seta on a low protuberance; coxae II lacks anterodorsal spines; coxae lack ventral spurs or spur-like spines and ridges. Chaetotaxy of legs: I= 0 0/1 0/1 0 (2), 1 0/2 1/1 1 (6), 2 3/1 2/3 2 (13), 2 3/2 3/1 2 (13), 2 3/2 3/1 2 (13), 4 8/4 2/2 6/3 4 (33) + 6 solenidia + 1 macroseta; II= 0 0/1 0/1 0 (2), 1 0/2 0/1 1 (5), 2 3/1 2/2 1 (11), 2 3/1 2/1 2 (11), 2 2/1 2/1 2 (10), 3 3/2 1/1 3/2 3 (18); III= 0 0/1 0/1 0 (2), 1 1/2 0/1 0 (5), 1 2/1 1/0 1 (6), 2 2/1 2/1 2 (10), 2 1/1 2/1 2 (9), 3 3/2 1/1 3/2 3 (18) and IV= 0 0/1 0/0 0 (1), 1 1/2 0/1 0 (5), 0 2/2 1/0 1 (6), 2 2/1 3/1 1 (10), 2 1/1 3/1 2 (10), 3 3/2 1/1 3/2 3 (18). Sensory field of tarsus I has distal setae (d1–d2) on pedicels; setae s4 (solenidion) fused at base; two setae (solenidion) additional in tarsus (“s?”), a seta near ds and d2, and a very posterior seta; one macroseta, long and slender with distinctly wide socket on dorsal (seta “m”, probable).

Gnathosoma — (Figure 4A–F). Deutosternal groove with nine denticles arranged 1–1–

Figure 3 *Chiasmanyssus cavernicola* n. sp. female. A – Dorsal shield; B – Tritosternum; C – Sternal shield, note the distinct X-shaped sclerotization and epigynal shield; D – Anal shield. Scale bars: A 20 μ m; B, C & D 50 μ m.

Figure 4 *Chiasmanyssus cavernicola* n. sp. female. A – Gnathosomatic dorsal; B – Gnathosomatic ventral; C – Venter palpal genu with distinct circular (spatulate) and membranous seta; D – Dorsal palpal genu with lyrifissure; E – Palpal trochanter with ventral process; F – Chelicera. Scale bars: 50 μm.

Figure 5 *Chiasmanyssus cavernicola* n. sp. male. A – Idiosomatic dorsum; B – Idiosomatic ventral. Scale bars: 100 µm.

1–1–1–1–1–1–1. Gnathosoma with three pairs of hypostomal setae (hyp1, hyp2 and hyp3) slender and subequal and one pair of gnathosomal setae (pc). Trochanter–tarsus chaetotaxy formula 0/2 (2); 1 1/0 1/0 1/0 1 (5); 2 1/0 1/0 1/0 1 (6); 2 2/1 2/0 3/1 3 (14) and tarsus with 16 setae + apotele forked. Disto-ventral with circular (spatulate) and membranous seta (Figure 4C) and one pair of dorsal lyrifissure on posterior margin of palpal genu (Figure 4D). Palpal trochanter has a small blade-like ventral process, not extending beyond the segment (Figure 4E). Chelicera with chelae simple and slender, without spines or other process (Figure 4F).

Description. Male — Measurements summarized in table 4 (Figures 5–7)

Idiosoma — (Figure 5A–B). As in female but smaller. *Dorsum* (Figure 5A). Dorsal shield entire, occupying most of the dorsal idiosoma; anterior margin of the dorsal shield not discernible from the soft cuticle; posterior narrow to setae J5; reticulate sculpture; dorsal shield with j and J series complete, j1, j2 and j3 on dorsal shield and J5 minute, bears approximately 56 pairs of setae; neotrichous area on opisthonthal region; a pair of additional minute setae,

Figure 6 *Chiasmanyssus cavernicola* n. sp. male. A–C: Legs II, III and IV with chaetotaxy of setae dorsal and ventral (black). Scale bars: A 20 µm; B & C 50 µm.

Figure 7 *Chiasmanyssus cavernicola* n. sp. male. A – Leg I; B – Chaetotaxy of setae in tarsus I; C – Tarsus I with setae in sensillary field; D – Chelicera. Scale bars: A, B & C 50 µm; D 20 µm.

laterally to J4 and J5, on neutrichous area of opistonotal region; one pair of lyrifissures near on margin lateral to setae j1 and six pairs of pores visible on dorsal shield, four pairs of conspicuous pores near setae j3, z4, J3, one laterally to J4 and J5 setae, and two supplementary pairs positioned close together, in the dorsal shield margin, laterally the setae J2; four pairs of caudal setae on unarmed dorsum. *Venter* (Figure 5B). Tritosternum as in female. Venter has a holovertral shield; reticulated sculptured; bears two pairs of lyrifissures and three pairs of setae on sternal region; third pair of pores near the metasternal and genital setae on shield; ventral region broadened posteriorly, with 10–11 setae on each side. Anal region with three post-anal setae, cribrum wide and anal ring present. Endopodal plates small and triangular, between coxae III and IV. Unarmed ventre with four pairs of setae. Peritreme much shorter than in female, extending from posterior margin of coxa II to anterior margin of coxa IV. Peritrematic shield anteriorly fused to dorsal shield, with nodule fused to the middle of the shield. Peritrematic shield posterior to stigma as in females.

Legs (Figure 6A–C and Figure 7A–C). Legs as in female. Chaetotaxy of legs as in female, including tarsus I sensory field (Figure 7B–C).

Gnathosoma — As in female. Palpal trochanter lacks a ventral process and chelicera with spermatodactyl short (Figure 7D).

Description. Protonymph — Measurements summarized in table 4. (Figures 8–9).

Idiosoma — (Figure 8A–B). Smaller than adults. *Dorsum* (Figure 8A): Subrectangular podonotal shield with lateral borders biconcave at level of z2 and z4, posterior region almost square; some line sculpture on dorsal shield surface; 11 pairs of setae, with j2 very close to j1; two pairs of pores near setae j3 and s5 on podonotal shield. Dorsum with two pairs of medial setae between the shields (J1–J2) and a lateral pair (J3). Mesonotal shield (or median diastema or platelets) comprising three pairs of platelets. Pygidial shield trapezoidal, as long as wide, with pairs of setae: J4–J5, Z3–Z5 and S5; J4 and Z4 much smaller than other setae (except J5, minute) on the pygidial shield; three pairs of pores near setae Z3, Z4 and J5 on pygidial shield. Unarmed dorsum has three pairs of free “nodules”, two pairs large near to lateral edges and one pair very small on anterior region. Unarmed dorsum with 12 pairs of long setae, dorsal and ventral caudal setae may be present 1 or 2 minute barbs (clearly observed just under immersion). *Venter* (Figure 8B). Tritosternum as in adults. Sternal shield resembles a pentagon; three pairs of slender and long sternal setae; two pairs of lyrifissures posterior to st1 and st2. Unarmed venter with five pairs of setae, four pairs of median setae between ventral shields and a pair of caudal setae (without seta flanking the anus); setae pair between coxae IV are slender than other ventral setae. Anal shield subtriangular with postanal setae long and subequal; Peritreme short, extending from middle of coxae III to middle of coxae IV.

Legs — (Figure 9A–D). Chaetotaxy of the legs: I= 0 0/1 0/1 0 (2), 1 0/2 0/0 1 (4), 2 3/1 1/1 2 (10), 1 2/1 2/1 1 (8), 1 2/1 2/1 1 (8), 4 6/3 1/1 6/3 3 (27) + 6 solenidia+ 1 macroseta; II= 0 0/1 0/1 0 (2), 1 0/1 0/1 1 (4), 1 2/1 2/1 1 (8), 1 2/0 2/0 1 (6), 1 1/1 2/1 1 (7), 3 3/2 1/0 3/2 3 (17); III= 0 0/1 0/1 0 (2), 1 0/1 0/1 1 (4), 1 2/1 1/0 0 (5), 1 2/0 2/0 1 (6), 1 1/1 2/1 1 (7), 3 3/2 1/0 3/2 3 (17) and IV= 0 0/1 0/0 0 (1), 1 1/2 0/0 0 (4), 0 2/1 1/0 0 (4), 1 2/0 2/0 0 (5), 1 1/1 2/1 1 (7), 3 3/2 1/0 3/2 3 (17). Tarsus I sensory field has setae as in adults.

Gnathosoma — The gnathosoma, including chelicerae, similar to females, except the trochanter–tarsus palp chaetotaxy formula: 0/1 (1); 1 0/0 1/0 1/0 1 (4); 1 1/0 1/0 1/0 1 (5); 1 4/1 4/1 1 (12) and tarsus with 15 setae + forked apotele. Palpal trochanter lacks a ventral process.

Deutonymph and Larva — Unknown.

Host — Unknown

Etymology — The specific name “*cavernicola*” refers to its habitat.

Material studied — *Holotype*: Female [UFMG AC 171121], on 12-26/Aug./2013 at 18°56'10.6"S 43°24'45.5"W, in cave **CSS-09**, Conceição do Mato Dentro municipality. *Paratypes*: Female [UFMG AC 180094], on 26/Jun- 06/Jul./2018, at 19°20'35.0"S 43°18'28.8"W, in cave **MCFC-0012**, Itambé do Mato Dentro municipality; Female [UFMG AC AC171375], on 21/Nov. and 02/Dec./2016, in cave **HOLC-0090/97**, at 19°36'10.5"S

Figure 8 *Chiasmanyssus cavernicola* n. sp. protonymph. A – Idiosomatic dorsum; B – Idiosomatic venter. Scale bars: 50 μ m.

44°00'43.1"W, Pedro Leopoldo municipality; Female [UFMG AC 180044], on 26-28/Mar./2018, at 20°12'07.8"S 43°58'03.7"W, in cave **MS-0030**, Nova Lima municipality; Female [UFMG AC 170277], on 16-26/Jan./2017 at 19°09'58.7"S 43°15'59.3"W, in cave **SPT-0180**, Conceição do Mato Dentro municipality; Three female [UFMG AC 160229, 160327, 160214] and one male [UFMG AC 160322], on 10-27/Apr./2015, at 16°08'56.0"S 44°37'39.0"W, in cave **Lapa Sem Fim**, Luizlândia municipality; Female [UFMG AC 180121], on 26/Jun. and 06/Jul./2018, at 19°19'52.2"S 43°18'41.8"W, in cave **MCFC-0009**, Itambé do Mato Dentro municipality; Female [UFMG AC 200027], male [UFMG AC 180341] and protonymph [UFMG AC 200032], on 30/Apr. and 05/May/2018, at 18°55'08.0"S 43°25'13.6"W, in cave **CSS-0007**, Conceição do Mato Dentro municipality; Protonymph [UFMG AC 180049], on 26-28/Mar./2018, 20°12'08.7"S 43°58'03.4"W, in cave **MS-0031**, Nova Lima municipality. All localities in Minas Gerais state, Brazil. Coordinates given in WGS-84 All mites were collected freely dwelling on cave floor by the staff of the company Carste Ciência e Meio Ambiente. The additional material analyzed can be seen in the supplementary material 1.

Figure 9 *Chiasmanyssus cavernicola* n. sp. protonymph. A–D: Legs I, II, III and IV with chaetotaxy of setae dorsal and ventral (black). Scale bars: A & B 50 μ m; C & D 20 μ m.

Discussion — The new genus differs from *Synasponyissus*, *Bewsiella* Domrow 1958, *Glauconyssus* Uchikawa 1991, *Ichoronyssus* Kolenati 1858 and *Parichoronyssus* by bearing less than three pairs of pores on the sternal shield. These genera have the third pair of sternal pores on the shield or as in *Synasponyissus* on the complex sternal-median shield, included in the so-called group “A” by Radovsky (2010).

Chiasmanyssus cavernicola **n. gen. n. sp.** resembles all other genera of Macronyssidae by sharing the peritrematic shield posterior to the stigma fused to the parapodal shield around the posterior margin of coxa IV in adults, a character regarded as apomorphic in the family, but absent in several genera (*Bewsiella*, *Glauconyssus*, *Ichoronyssus*, *Synasponyissus* and *Parichoronyssus*) probably close to the laelapine stem (Radovsky 2010), and by the presence of an extra seta in the tibia III, genu III and IV.

Those setae are most likely lost in some derived genera: i) all extra seta absent from *Chiroecetes*; ii) only extra setae on genu III in *Megistonyssus* Radovsky 1966; iii) tibia III and genu IV both with one extra seta in *Mitonyssoides* and *Acanthonyssus*; iv) only genu IV with one extra seta in *Macronyssus* and *Radfordiella* (one species *R. oudemansi* Fonseca 1948); v) *Pellonyssus*, *Atrichonyssus* Radovsky 2010 and *Ophionyssus* have two extra setae in genu IV. Radovsky (2010) suggests that absence or presence of one or more extra setae on any or all tibia III, genu III and IV is useful for setting apart closely related genera within the family.

Chiasmanyssus **n. gen.** differs from all other genus described by the morphology of the sternal shield in females with its distinct “X” shaped area. Some genera have the sternal shield with some distinct sclerotization but not comparable to this: *Chelanyssus* Zumpt and Till 1953 and *Steatonyssus* have a sclerotized band in posterior margin (Till and Evans 1964; Till and Evans 1966; Radovsky 1966); *Chiroptonyssus* has a heavily sclerotized posterior band extending anteriorly on the sides of the shield and *Lagidonyssus* has the shield divided into two parts, which anterior part shortest and a posterior part (from first pair of pores) more strongly sclerotized and with setae st2 and st3 (Fonseca 1935b; Radovsky 1967; 2010).

The presence of the seta J5 minute is similar to *Macronyssus* and *Megistonyssus*, among the most derived genera. However, it is distinguished from these genera by the absence of anterior spur of coxa II. In *Macronyssus*, the protonymphs have only 10 setae in shield podonotal, with j1 setae are anterior to the podonotal shield and in *Megistonyssus* the pygidial shield has only four setae pairs and a setae pair flanking the anal shield in the unarmed venter (Radovsky 1966; 1967; 2010).

Chiasmanyssus **n. gen.** is closely related to genus *Cryptonyssus* Radovsky 1966. It can be set apart, however, by only five pairs of setae on venter, four between the shields and one caudal (without setae flanking the anus); shape of pygidial and podonotal shield and presence of only 12 setae pairs on the dorsal integument. Furthermore, females lack marginal setae on dorsal shield, while males lack stout or spinose anterior setae on coxae II and III (Radovsky 1966; 1967).

Despite the scanty molecular data that could be retrieved from GenBank, the 16S fragment sequenced apparently is a good bar-coding marker, since it has a considerable gap between intra- and inter-specific distances. This fragment also supported *Chiasmanyssus* **n. gen.** as an independent lineage, highly supported in a clade recovered as sister-group of *Ornithonyssus* and *Lepronyssoides*.

Finally, it remains as a mystery the host of the new species. As the large number of individuals dwelling on caves floor, vertebrates that use them as nesting sites or shelter are candidates. However, authors have been surveying bats for several months, the most abundant vertebrates found in those caves and, despite finding most of mesostigmata lineages usually associated to bats in neotropics, could not find *Chiasmanyssus* **n. gen.** on them thus far.

Keys for genus of Macronyssidae from Brazil

Females

1. Peritrematic shield posterior to the stigma not fused with the parapodal shield around the posterior margin of coxa IV; three pairs of pores on sternal shield; setae z3 on dorsum; setae J5 absent; dorsal shield with lateral and posterior portions darkened and submerged beneath striated cuticula; cinctures absent in chelae *Parichoronyssus*
 — Peritrematic shield posterior to the stigma fused to the parapodal shield around the posterior margin of coxa IV; less than three pores pairs on sternal shield; setae z3 absent; setae J5 present or absent; dorsal shield without portions submerged; cinctures may be present in chelae ... (2)
2. Genu III with 9 setae; cinctures may be present on chelae (3)
 — Genu III with 10 setae; cinctures absent on chelae (5)
3. Ventral process of palpal trochanter absent; cinctures absent on chelae; coxa II without anterior spur; tibia III with nine setae *Mitonyssoides*
 — Ventral process of palpal trochanter present; cinctures present on chelae; coxa II with anterior spur; tibia III with less than nine setae (4)
4. Setae J5 absent; sternal glands absent on sternal shield; accessory setae absent on epigynal shield; ventral shields lack sculpturing *Radfordiella*
 — Setae J5 present and minute; sternal glands present, lateral to the first pair of pores and the glands are variable in structure on shield; accessory setae present or absent on epigynal shield; ventral shields with some distinct sculpturing *Macronyssus*
5. Epigynal shield flanked by the genital setae inserted on the unarmed integument; chelicera basally swollen *Ophionyssus*
 — Epigynal shield with genital setae inserted on shield; chelicera basally not as above (6)
6. Some or most dorsal and caudal setae of idiosoma barbed (7)
 — Barbed setae absent on idiosoma (or at most 1–2 pairs of caudal setae notched or feathered) (10)
7. Ventral process of palpal trochanter absent; sternal shield with less than 3 pairs of setae *Thigmonyssus*
 — Ventral process of palpal trochanter present; sternal shield with 3 pairs of setae (8)
8. Setae J3 present; J5 usually present (absent in *C. cubensis* Dusbábek 1969)]; some caudal setae are peg-like and with tip multiply barbed; setae on the legs smooth; sternal shield with a heavily sclerotized posterior band extending anteriorly on the sides of shield; claw I much smaller than the others claws; leg II stout than the others legs *Chiroptonyssus*
 — Setae J3 and J5 absent; caudal setae are not peg-like and with single barbed tip; setae on the legs smooth or barbed; sternal shield lacking a sclerotized band; subequal claws and legs in width (9)
9. Setae j3 present; all setae on legs smooth; sternal shield with smooth oval structures posterior to st1 and posteromedian to the first pair of pores; always three pairs of setae on sternal shield; coxa III with a posterior triangular, round-tipped spur *Lepronyssoides*
 — Setae j3 absent; usually legs with barbed setae, but a single barb; sternal shield lacking oval structure; usually three pairs of setae on sternal shield, rarely st3 are off the shield (some *O. sylviarum* and *O. hypertrichus* Radovsky 2007); coxa III with a setiform posterior seta *Ornithonyssus*

10. Dorsal shield divided (11)
 — Dorsal shield entire (12)
11. Setae j3 and J5 absent; 1–2 pores pairs on sternal shield; sternal shield lacking sclerotized band; genu IV with two extra setae (11 setae) *Pellonyssus*
 — Setae j3 usually present (absent in e.g. *S. evansi* Delfinado 1960, *S. benoiti* Till and Evans 1964), often much reduced; J5 usually present, rarely absent; two pores pairs on sternal shield; sternal shield with a sclerotized band in posterior margin; genu IV with one extra setae (10 setae) *Steatonyssus*
12. Setae J5 absent; dorsal shield always non-neotrichous; some coxae with ridges; claws subequal *Macronyssoides*
 — Setae J5 present; dorsal shield may be neotrichous; coxae without ridges; claws may be unequal (13)
13. Dorsal shield neotrichous; setae J5 small, subequal to Z5; most 1–2 pairs of caudal setae notched or feathered; sternal shield lacking distinct sclerotization; without an anterior protusion on epigynal shield; claw I reduced relative to other claws *Mitonyssus*
 — Dorsal shield non-neotrichous; setae J5 minute; none of caudal setae notched or feathered; sternal shield with distinct X-shaped area; anterior edge of epigynal shield with a protusion; claws subequal *Chiasmanyssus* **n. gen.**

Males

1. Peritrematic shield posterior to the stigma not fused, without the parapodal shield around the posterior margin of coxa IV; tarsus I with 2–4 strongly pedicellate distal setae; setae J5 absent; tibia III, genu III and IV without extra setae *Parichoronyssus*
 — Peritrematic shield posterior to the stigma fused, with the parapodal shield around the posterior margin of coxa IV; tarsus I with 0–1 pedicellate distal setae setae J5 present or absent; some tibia III, genu III and IV with extra setae (2)
2. Genu III with 9 setae (3)
 — Genu III with 10 setae (5)
3. Setae st1 arise in the presternal area; neotrichous on dorsal shield *Mitonyssoides*
 — Setae st1 arise in the esternal shield; non-neotrichous on dorsal shield (4)
4. Setae pair J5 present and minute; ventral process of palpal trochanter with a strong longitudinal ridge; dorsal and/or caudal margin may bear some setae long and flagelliform; fixed chela with a tiny pilus dentilis; leg I slender than or equal to the other legs; all claws small; coxa II with anterior spur entire (except *M. constrictus* with bifid tip) *Macronyssus*
 — Setae pair J5 absent; without ventral process of palpal trochanter or at most with a line showing its position; dorsal and/or caudal margin never flagelliform; fixed chela without a tiny pilus dentilis; leg I and II stout but leg II may be slightly to much stronger than the other legs; all claws are strong; coxa II with anterior spur bifid or 2 subequal spur *Radfordiella*
5. Some setae barbed on idiosoma at least caudally with 1–3 barbed setae (6)
 — All setae smooth (or 1–2 pairs of caudal setae notched or feathered) on idiosoma (9)
6. Setae j1 and j2 off dorsal shield *Thigmonyssus*
 — Setae j1 and j2 on dorsal shield (7)

7. Palpal femur without enlarged lateral setae and one pedicel; J5 usually present (absent *C. cubensis*, small); venter usually has sternogenital and ventrianal shields or rarely holoventral shield *Chiroptonyssus*
— Palpal femur with spinose seta and on a lateral pedicel; J5 absent; venter always has a holoventral shield (8)
8. Setae j3 absent; setae in the opisthonotal region, not including neotrichae behind J2; 1 or 2 enlarged lateral setae on palp femur, of which the anterior one on a prominent pedicel; all legs with some dorsal setae barbed *Ornithonyssus*
— Setae j3 present; setae in the opisthonotal region, including about 3–4 neotrichae behind J2; palpal femur has only 1 enlarged lateral seta on pedicel; legs setae lack barbs *Lepronyssoides*
9. Venter with sternogenital and anal shields, the ventral region unarmed; genital setae inserted on the unarmed integument flanking the sternogenital shield; Chelicera is basally swollen as in the female *Ophionyssus*
— Venter with a holoventral shield or sternogenital and ventrianal shields; genital setae on the shield; Chelicera basally not as above (10)
10. 1–2 pairs of caudal setae notched or feathered; claws I greatly reduced relative to other claws *Mitonyssus*
— None setae as above; claws I not reduced relative to other claws (11)
11. Genu IV with 2 extra setae (11 setae) *Pellonyssus*
— Genu IV with 1 extra seta (10 setae) (12)
12. Setae J5 absent; some setae on dorsal shield with expanded tip *Macronyssoides*
— Setae J5 present or absent; none setae on dorsal shield with expanded tip (13)
13. Setae J5 minute; j3 subequal to other setae on shield; anterior spur in coxa II absent; coxa III has an anterior seta setiform *Chiasmanyssus* **n. gen.**
— Setae J5 when present small, not minute; setae j3 very reduced or absent; anterior spur in coxa II present with tip divided into several denticles; coxa III has an anterior seta ranging from normal setiform to regular spinose to spinose with a blade-like proximal expansion *Steatonyssus*

Protonymphs

1. Setae j1 anterior to podonotal shield or may be on margin of podonotal shield (2)
— Setae j1 on podonotal shield (4)
2. Setae j2 off podonotal shield *Thigmonyssus*
— Setae j2 on podonotal shield (3)
3. Podonotal shield with 10 setae pairs; setae j3 present; J5 present and minute; pygidial shield with 6–7 setae pairs; 2 median setae between dorsal shield; normally 4 setae pairs between ventral shields but may be more than 10 setae pairs when neotrichous *Macronyssus*
— Podonotal shield with 8–9 setal pairs; setae j3 absent; J5 absent; pygidial shield with 1–3 setae pairs; 3 median setae between dorsal shield; 2–3 setae pairs between ventral shields, never neotrichous *Pellonyssus*
4. Usually no more than two median setal pairs between dorsal shields, a posterior pair (J3) may be present in integument, in which case it is shifted laterally (5)

- About 3–8 or more median setal pairs between dorsal shields, setae J3 pair when present on integument, not laterally shifted (9)
- 5. Chelicera with a swollen base; on tarsus I, seta s2 small much less than half the length of seta s1 *Ophionyssus*
- Chelicera with an approximately uniform base in width throughout its length; seta s2 on tarsus I not as above (6)
- 6. Setae J5 present and minute; pygidial shield with six setae pairs *Chiasmanyssus* **n. gen.**
- Setae J5 absent; pygidial shield no more than four setae pairs (7)
- 7. Pygidial shield with two setae pairs; sensory field of tarsus 1 with 2–4 pedicillate distal setae; genital setae pair absent; two pairs flanking the anal shield; none extra seta on tibia III, genu III and IV *Parichoronyssus*
- Pygidial shield with 3–4 setae pairs; sensory field of tarsus 1 with 0–1 pedicillate distal setae; genital setae pair present; 0–1 pairs flanking the anal shield; all tibia III, genu III and IV with one extra seta or at least genu IV with an extra seta (8)
- 8. Setae j3 present, may be reduced but always represented by the trichopores; pygidial shield with a pair of enlarged anterolateral pores; pygidial shield normal or much reduced; none extra seta in tibia III and genu III but genu IV may be with one extra seta; claw I not larger than other legs; peritreme lacking a post-stigmal part *Radfordiella*
- Setae j3 present and subequal other shield setae; pygidial shield lacking enlarged pores; pygidial shield always normal; one extra seta on tibia III, genu III and IV; claw I larger than those of the other legs; peritreme with a post-stigmal part *Macronyssoides*
- 9. Three median setal pairs between dorsal shields (non-neotrichous) (10)
- Four or more median setal pairs between dorsal shields (neotrichous) (12)
- 10. All setae smooth on idiosoma *Steatonyssus*
- Barbed setae on idiosoma, usually single barb (11)
- 11. Podonotal shield with 10 pairs of setae; j3 absent; Z4 not reduced; all legs with some barbed setae *Ornithonyssus*
- Podonotal shield with 11 pairs of setae; j3 present; Z4 reduced; all legs with smooth setae *Lepronyssoides*
- 12. Venter with only 3 pairs of setae between the ventral shield *Mitonyssoides*
- Venter with more than 3 pairs of setae between the ventral shields (13)
- 13. Venter neotrichous with about 5 or 6 pairs of setae between shields (except *C. venezolanus*); some caudal setae on the pygidial shield peg-like and/or with short barbs; leg II much stouter than other legs; claws II much larger than other claws and claws I as claws III and IV *Chiroptonyssus*
- Venter neotrichous with about 8–11 pairs of setae between shields; some caudal setae on the pygidial shield; leg II with normal thickness as the other legs; claws II subequal other claws but claws I much reduced or absent *Mitonyssus*

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