

A new genus and species for the first recorded cave-dwelling Cavernicola (Platyhelminthes) from South America

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Abstract

Species diversity of Brazilian cave fauna has been seriously underestimated. A karst area located in Felipe Guerra, northeastern Brazil, which is a hotspot of subterranean diversity in Brazil, has revealed more than 20 troglobitic species, most of them still undescribed. Based on recent samplings in this karst area, we document the occurrence of the suborder Cavernicola (Platyhelminthes) in South American hypogean environments for the first time and describe a new genus and species for this suborder. *Hausera* Leal-Zanchet & Souza, **gen. n.** has features concordant with those defined for the family Dimarcusidae. The new genus is characterized by two unique features, viz. an intestine extending dorsally to the brain and ovovitelline ducts located dorsally to the nerve cords, which is complemented by a combination of other characters. The type-specimens of *Hausera hauseri* Leal-Zanchet & Souza, **sp. n.** are typical stygobionts, unpigmented and eyeless, and they may constitute an oceanic relict as is the case of other stygobiotic invertebrates found in this karst area in northeastern Brazil.

Keywords

Troglobitic species, subterranean diversity, freshwater flatworms, triclads

Introduction

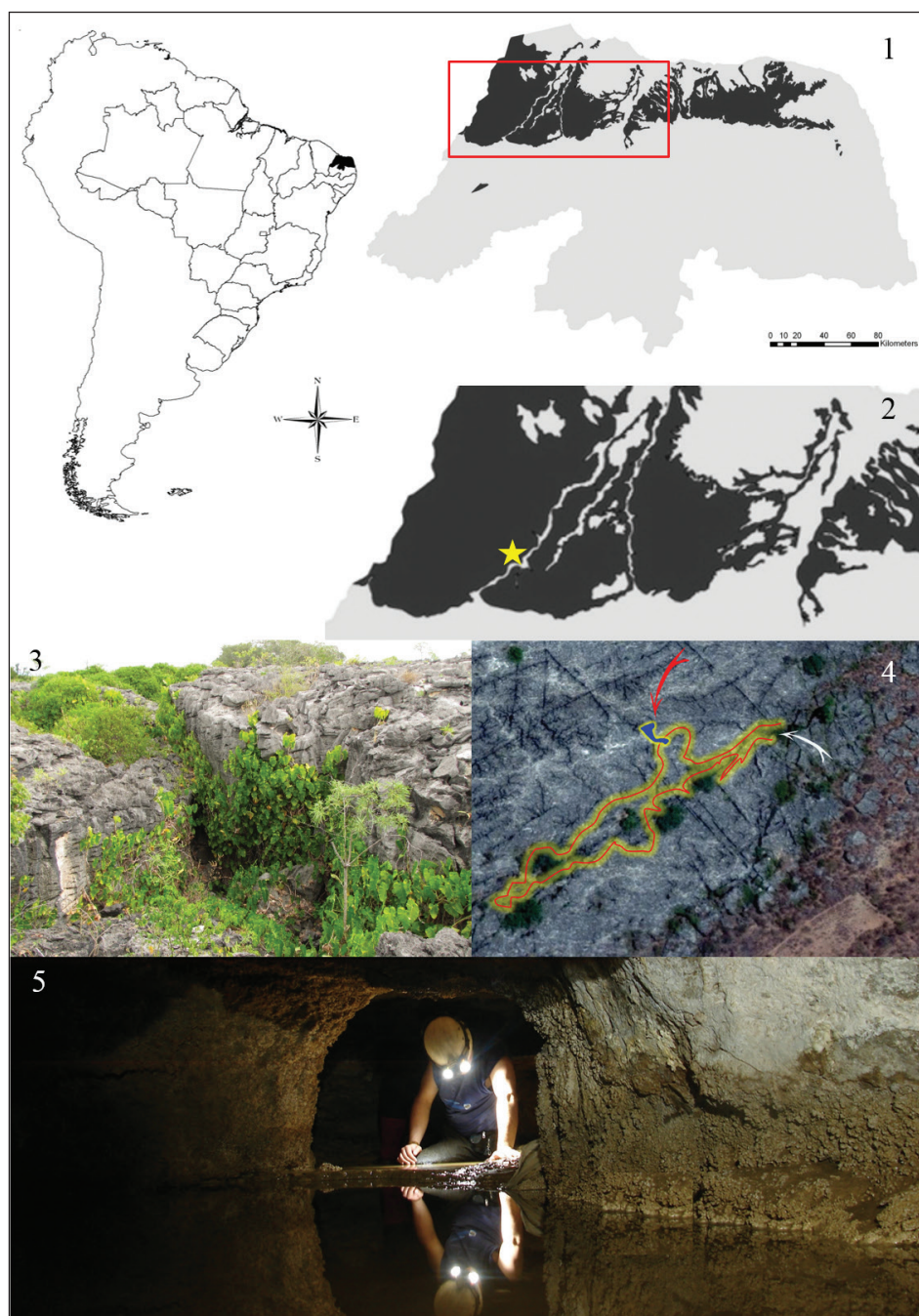
Species diversity of Brazilian cave fauna has been seriously underestimated (Ferreira 2005). Although more than 11,000 caves have been documented in Brazil, this may represent only 10% of the potential number of caves in the country (Auler et al. 2001). Studies performed in unexplored areas have revealed dozens of new troglobitic taxa, some of which were only recently described (Pellegrini and Ferreira 2014, Azara and Ferreira 2013, Iniesta and Ferreira 2013). One of the undescribed species, sampled in a karst area in northeastern Brazil, is a freshwater flatworm hereby assigned to the suborder Cavernicola (Platyhelminthes: Tricladida).

The triclad infraorder Cavernicola Sluys, 1990 was proposed to encompass five species of the family Dimarcusidae Mitchell & Kawakatsu, 1972 and now constitutes one of the three triclad suborders (Sluys et al. 2009). This taxon includes four genera, viz. *Rhodax* Marcus, 1946, *Opisthobursa* Benazzi, 1972, *Balliania* Gourbalt, 1972 and *Novomitchellia* Özdikmen, 2010 (Sluys 1990, Sluys et al. 2009, Özdikmen 2010). As its name indicates, most species of Cavernicola have type-localities in speleological habitats; they were recorded in Mexico, Tahiti Island and East Malaysia, respectively (Kawakatsu and Mitchell 1984, Sluys 1990). *Rhodax evelinae* Marcus, 1946 is an exception because its type-locality is in surface water in São Paulo city, southeast Brazil (Marcus 1946). Asexual specimens of presumed *Rhodax* were also recorded in surface water from southern Brazil (Braccini and Leal-Zanchet 2013, Vara and Leal-Zanchet 2013), and they have been introduced in tanks of tropical fishes in Japan (Kawakatsu et al. 1985, 1995, Sluys et al. 2010). In addition, the occurrence of a morphospecies of presumed *Opisthobursa* was reported in a hypogean environment in Guatemala after a morphological analysis of immature specimens (Kawakatsu and Mitchell 1983). Thus, the diversity of the suborder Cavernicola in South-American subterranean habitats is largely unknown.

After erecting the taxon Cavernicola and since the description of *Novomitchellia sarakawana* (Kawakatsu & Chapman, 1983), no new species have been described for the family Dimarcusidae. In addition, the only known Cavernicola species from South America is *R. evelinae*, without records from subterranean habitats. Based on recent samplings in the Felipe Guerra karst area, northeastern Brazil, we provide here the first documentation of the family in South American hypogean environments by describing a new genus and species for this family.

Material and methods

Specimens were collected from the Crotes cave, a limestone outcrop located in Felipe Guerra (5°33'38.87"S; 37°39'31.80"W), Rio Grande do Norte, Brazil (Figs 1–3). The type-locality is situated in the Caatinga biome, which is dominated by a semi-arid climate (type BSw'h' of Köppen's classification). This cave is part of a complex of 70 caves called "Lagedo do Rosário", most of which have no perennial water bodies. The



Figures 1–5. Type-locality of *Hausera hauseri* Leal-Zanchet & Souza, sp. n.: **1–2** location in Rio Grande do Norte, Brazil, showing the range of limestone outcrops from the Apodi Group in detail (**2**) and the location of the Crotes cave (star) **3–4** surroundings of the main cave entrance (**3**) and aerial view of the region (**4**) indicating the cave contours (red/yellow line), main entrance (white arrow) and secondary gallery (red arrow); **5** perennial stream representing the sampling site.

Crotes cave was formed by the expansion of a diacclasis, which is a diagenetic fracture in the rock. The main gallery of this cave has a 237 m linear projection, and the roof has several vertical openings. Despite the dominance of dry substrates, a small perennial stream (Fig. 4) drains water from the epicarstic system. The flatworms were directly sampled from this water body (Fig. 5), which has a water depth of 10–15 cm.

During the field work, the specimens were photographed and fixed in ethanol 70%. Fixed specimens were analysed and photographed with a stereomicroscope. They were dehydrated and embedded in Paraplast. This material was sectioned at 5–7 μm and stained with hematoxyline/eosine or Goldner's Masson (Romeis 1989). Colour descriptors, based on the uptake of dyes of particular colours, were used for classifying secretions with trichrome methods: erythrophil, xanthophil and cyanophil. The term cyanophil is also applied to secretions that have an affinity for the green dye of Goldner's Masson.

Type-material was deposited in the following reference collections: Museu de Zoologia da Universidade do Vale do Rio dos Sinos, São Leopoldo, Rio Grande do Sul, Brazil (MZU), and the Helminthological Collection of Museu de Zoologia da Universidade de São Paulo, São Paulo, São Paulo State, Brazil (MZUSP).

Abbreviations used in figures

a: anterior tip; bc: bulbar cavity; br: brain; cg 1: cyanophil glands 1; cg 2: cyanophil glands 2; ci: cilia; cm: circular musculature; de: dorsal epidermis; eg: erythrophil glands; ej: ejaculatory duct; fgd: female genital duct; gic: genito-intestinal canal; i: intestine; in: insunk nuclei; lm: longitudinal musculature; m: mouth; ma: male atrium; o: ovary; ov: oviducts; pb: penis bulb; ph: pharynx; php: pharyngeal pouch; pp: penis papilla; r: rhabdites; rg: rhabditogen glands; s: sperm; sd: sperm duct; sg: shell glands; sv: seminal vesicle; t: testes; ve: ventral epidermis; vi: vitellaria; xg 1: xanthophil glands 1; xg 2: xanthophil glands 2.

Systematic account

Order Tricladida Lang, 1884

Suborder Cavernicola Sluys, 1990

Family Dimarcusidae Mitchell & Kawakatsu, 1972

Genus *Hausera* Leal-Zanchet & Souza, gen. n.

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Type-species. *Hausera hauseri* Leal-Zanchet & Souza, sp. n. Monotypic

Diagnosis. Dimarcusidae without eyes and without a copulatory bursa; female genital duct communicating with the intestine; ovovitelline ducts without caudal di-

chotomy, uniting to form a common ovovitelline duct; follicular testes; sperm ducts separately penetrating the penis bulb.

Distribution. Felipe Guerra (Crotes cave), Brazil

Etymology. The new genus is dedicated to the late Prof Dr Josef Hauser SJ as acknowledgement of his great enthusiasm for the study of freshwater flatworms. Gender: feminine.

Differentiation of the genus. The specimens of *H. hauseri* show features concordant with the definition of the family Dimarcusidae, viz. common oviduct or diverticulum oriented perpendicular to the horizontal bursal canal or female genital duct, penis bulb provided with glandular elements, ovaries generally located at some distance posterior to the brain, vasa deferentia (= sperm ducts) uniting to extra-bulbar common vas deferens or penetrating separately the penis bulb and testicular follicles fused or discrete (Sluys 1990). The specimens herein described show cell bodies of the penis glands within the bulb, the horizontal orientation of the female genital duct combined with the dorsal opening of the common oviduct, sperm ducts penetrating separately the penis bulb and discrete testicular follicles. The ovaries are situated posterior to the brain, but are close to it.

It is worth mentioning that the specimens of *H. hauseri* have a connection with the intestine that could be confused with a copulatory bursa in which the branch of the intestine immediately posterior to the bursa may stain differently from other parts of the posterior intestinal branches. In all examined specimens, the connections with other parts of the posterior intestinal branches could be traced, leading to the conclusion that a bursa is absent in *H. hauseri*.

Similarly to *Rhodax*, *Hausera* gen. n. does not have a copulatory bursa. Other diagnostic characters of *Rhodax*, however, such as presence of eyes, fused testes, sperm ducts uniting before penetrating the penis bulb and ovovitelline ducts with a caudal dichotomy do not occur in *Hausera* gen. n. Similarly to *Opisthobursa*, the sperm ducts separately penetrate the penis bulb in the new genus, but the latter lacks a bursa, in contrast to the genus *Opisthobursa*.

***Hausera hauseri* Leal-Zanchet & Souza, sp. n.**

<http://zoobank.org/44FA7072-366B-4967-91D6-C81E7FFE43E8>

Material examined. Holotype. MZUSP PL. 1562: coll. R. Ferreira, 22 March 2011, Crotes cave, Felipe Guerra, RN, Brazil – sagittal sections on five slides.

Paratypes. Crotes cave, Felipe Guerra, RN, Brazil. MZU PL.00142: coll. R. Ferreira, 22 March 2011 – sagittal sections on 10 slides; MZU PL.00148: coll. R. Ferreira, 22 March 2011 – transverse sections on 9 slides; MZU PL.00149: coll. R. Ferreira, 05 June 2010 – anterior region: transverse sections on 12 slides, posterior region: sagittal sections on 20 slides.

Diagnosis. *Hausera hauseri* can be distinguished from other species in the Dimarcusidae by (1) the numerous testicular follicles arranged in two irregular rows next to the margins of the body, extending from the level of the ovaries to the posterior end

Table 1. Measurements, in mm, of specimens of *Hausera hauseri* Leal-Zanchet & Souza, sp. n. after fixation. DG: distance of gonopore from anterior end; DM: distance of mouth from anterior end. The numbers given in parentheses represent the position relative to body length. * Measurements done after histological processing.

	Holotype MZUSP PL. 1562	Paratype MZU PL.00142	Paratype MZU PL.00148	Paratype MZU PL.00149
Length	4.5	7	5	7.5
Width	1.5	2	2	1.5
Length*	4.2	6.7	4.7	7.2
DM*	2.7 (64%)	5 (75%)	2.6 (55%)	5.5 (76%)
DG*	3.6 (86%)	5.5 (82%)	3.4 (72%)	6 (83%)

of the body; (2) the ovoid bulbar cavity with a posteriorly directed diverticulum into which the sperm ducts open; (3) a short and narrow transition region between the bulbar cavity and the ejaculatory duct; (4) the ovovitelline ducts arising from the lateral surface of the ovaries.

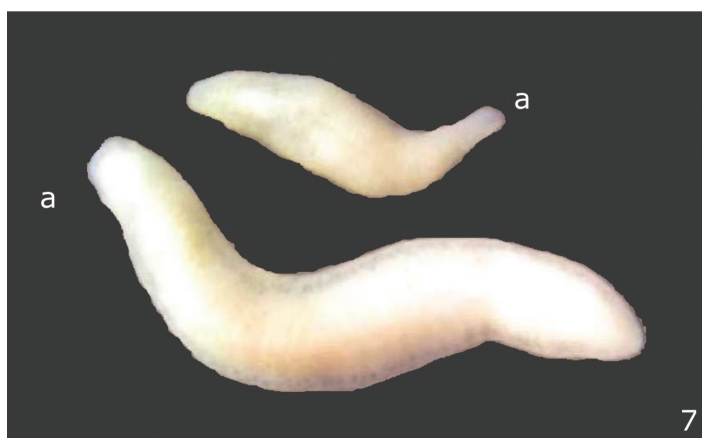
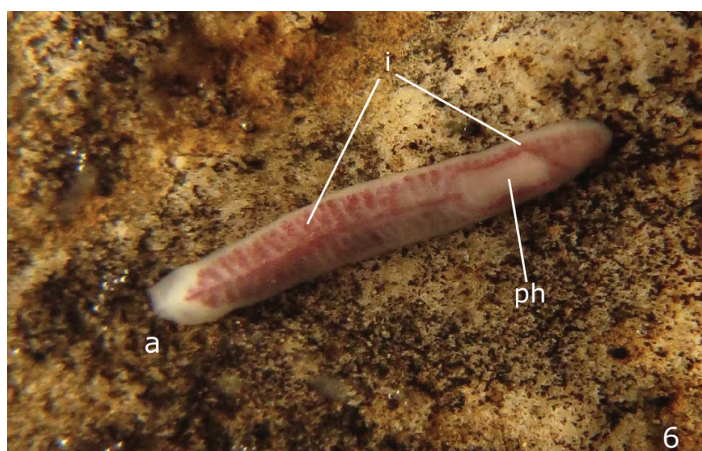
Description. Live specimens unpigmented and eyeless (Fig. 6). Head truncate; posterior tip pointed (Figs 6–7). Preserved specimens up to 7.5 mm long and 2 mm wide (Table 1). Mouth and gonopore located at the posterior body third (Table 1). Body margins almost parallel (Figs 6–8). After fixation, anterior and posterior tips rounded (Fig. 8).

Epidermis (Figs 9–10). Columnar, ciliated epithelium, with abundant xanthophil, rhabdogen secretion (rhammites), both dorsally and ventrally, being more densely distributed at the dorsal surface (Fig. 9). It also receives secretions of other four types of glands: (1) xanthophil, coarse granular secretion; (2) cyanophil amorphous secretion; (3) heavily stained cyanophil, fine granular secretion; (4) erythrophil, fine granular secretion (Figs 9–10). In addition, glands with heavily stained xanthophil, fine granular secretion open at the body margins and medially at the anterior and posterior tips of the body. Cilia more densely arranged on the ventral body surface (Fig. 10).

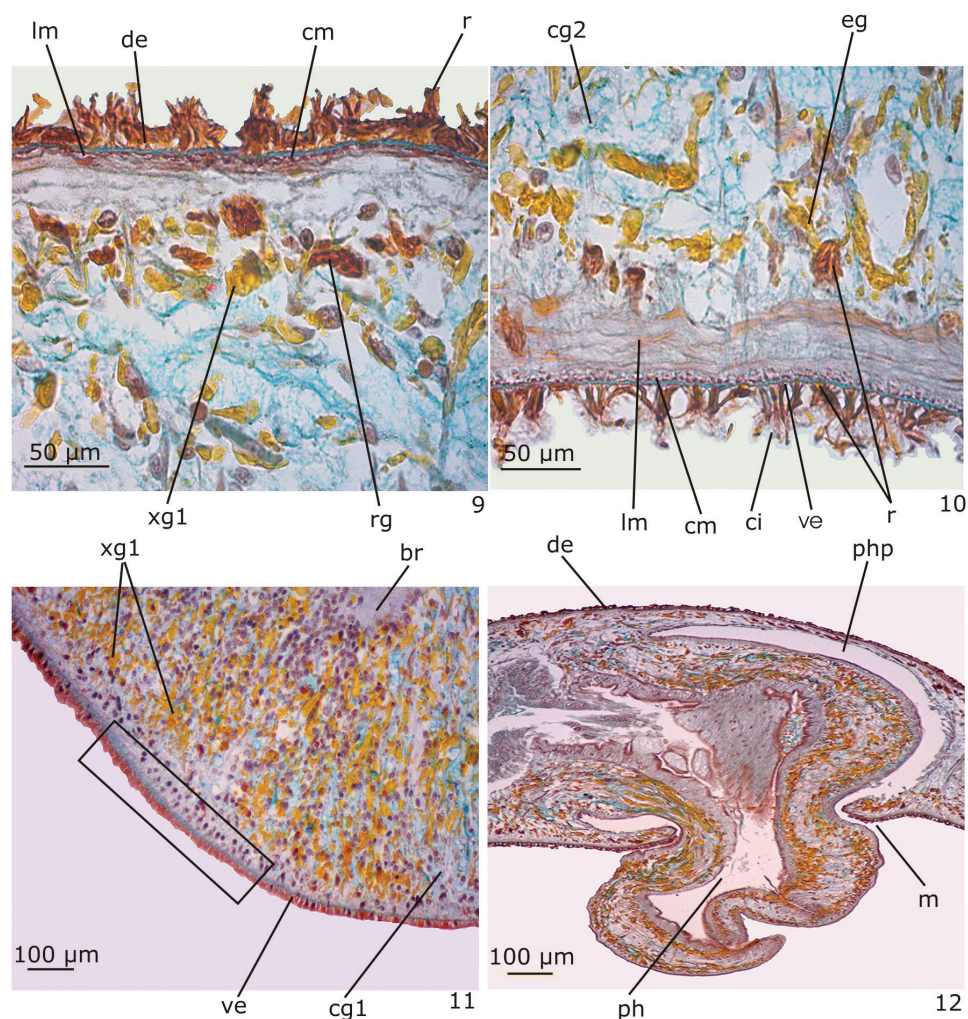
Cutaneous musculature (Figs 9–10). Two layers, viz. a thin subepithelial circular layer, followed by a thicker layer of longitudinal muscle. Dorsal and ventral cutaneous musculatures show similar height in the pre-pharyngeal region (13–15 μ m thick). In the anterior region of the body, the ventral musculature (35 μ m thick) is thicker than the dorsal one (9 μ m thick).

Sensory organs (Fig 11). Head with a pair of lateral sensory organs beginning about 140 μ m after the anterior tip. They are lined with densely ciliated columnar epithelium, highly innervated, with insunk nuclei and receive few openings of secretory cells. The cutaneous musculature is very thin at the level of the sensory organs.

Pharynx (Fig 12). Cylindrical; 0.9 mm long; it is located in the posterior third of the body. Pharynx and pharyngeal lumen lined with cuboidal ciliated epithelium with insunk nuclei. Three types of pharyngeal glands, viz. cells with coarse granular xanthophil secretion, cells with cyanophil amorphous secretion and less numerous cells with fine granular erythrophil secretion. Cell bodies of the pharyngeal glands located in the mesenchyme, mainly anterior and laterally to the pharynx. Outer musculature of the pharynx constituted of a thin subepithelial layer of longitudinal muscle, followed by



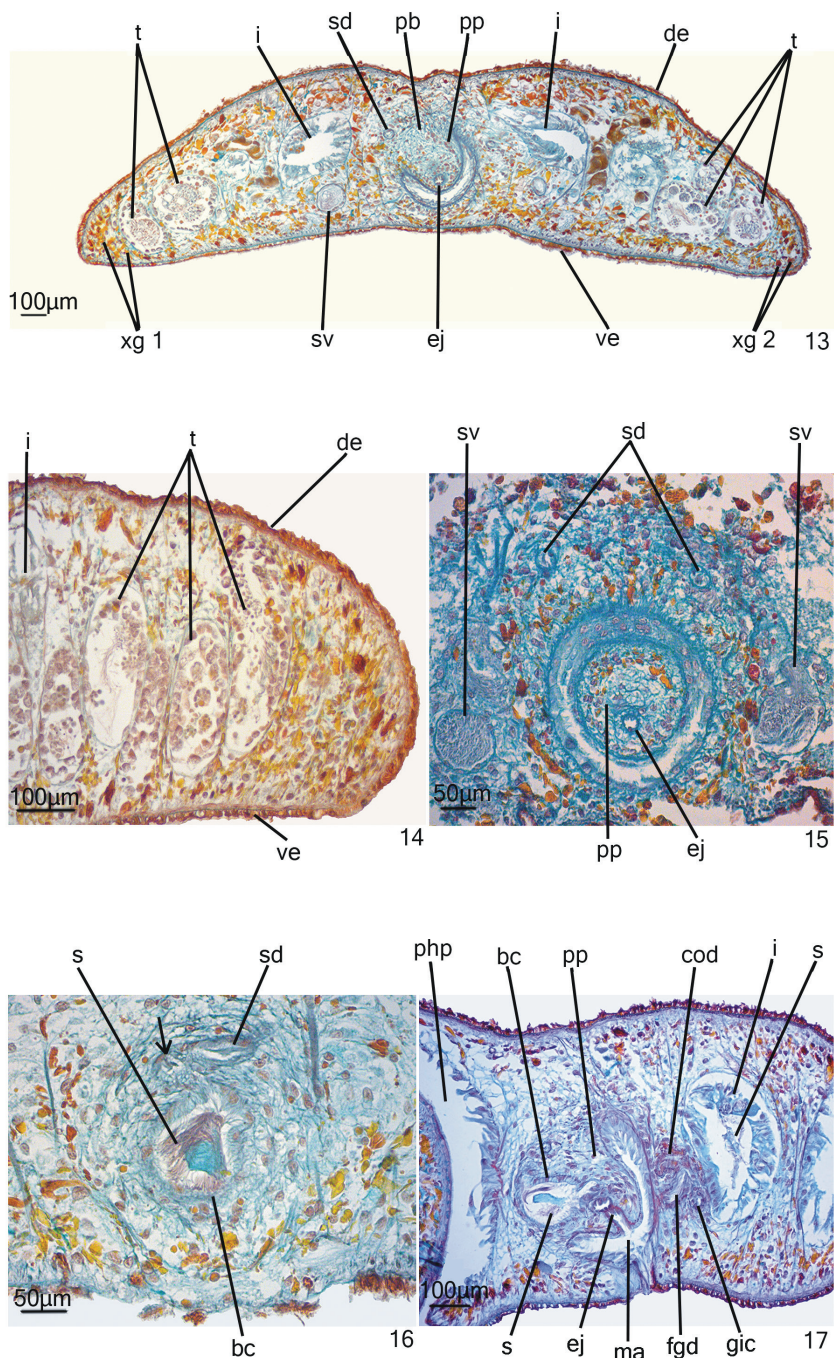
Figures 6–8. *Hausera hauseri* Leal-Zanchet & Souza, sp. n.: **6–7** photograph of live specimens in dorsal view, one of them (**6**) with intestine containing food and the pharynx visible **8** photograph of a preserved specimen (paratype MZU PL.00142) in dorsal view; grains of sand are seen over the dorsal surface. Scale bar for the figs **6–7** not available.



Figures 9–12. *Hausera hauseri* Leal-Zanchet & Souza, sp. n.: holotype (**9–10; 12**) and paratype MZU PL.00148 (**11**). **9–10** dorsal (**9**) and ventral (**10**) surfaces of the body in sagittal section **11** detail of the cephalic region in transverse section showing a sensory organ (rectangle) **12** sagittal section of the pharynx.

a thin layer of circular muscle, each about 5 µm thick. Inner pharyngeal musculature composed of a thick subepithelial layer of circular fibres (about 25–28 µm thick), followed by a layer of longitudinal fibres (about 10 µm thick).

Intestine (Figs 19, 22, 23). Anterior intestinal trunk extending onto the posterior part of the brain (Figs 19, 23). Posterior intestinal trunks anastomose and communicate with the reproductive system through a genito-intestinal duct (Fig. 22). The part of the intestine that meets the genito-intestinal duct is lined with a tall columnar epithelium composed of cells with cyanophil cytoplasm and cells with coarse granular,



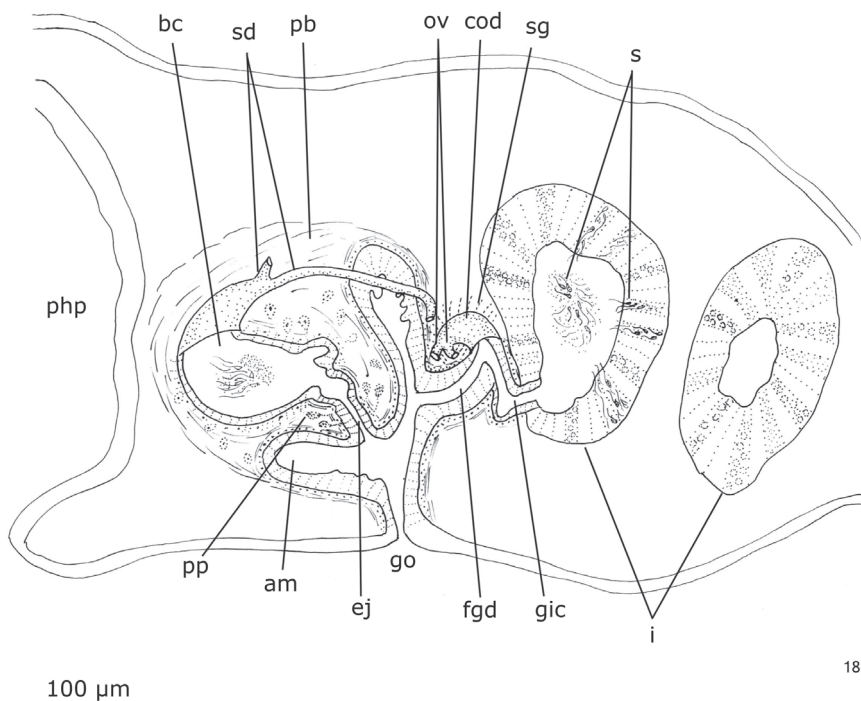
Figures 13–17. *Hausera hauseri* Leal-Zanchet & Souza, sp. n.: paratype MZU PL.00148 (**13; 15–16**) and holotype (**14; 17**) **13** transverse section of the body **14** sagittal section of the posterior tip **15–17** copulatory apparatus in transverse section (**15–16**) and sagittal section (**17**). The arrow indicates the diverticulum of the bulbar cavity.

erythrophil secretion; its lumen contains sperm and a small amount of erythrophil secretions (Fig. 22). Some spermatozooids are absorbed by the intestinal epithelium.

Male reproductive system (Figs 13–18). Numerous testicular follicles, 60–70 μm in diameter, arranged in approximately two irregular rows laterally to the intestinal diverticles, near body margins (Fig. 13). Testes extend from the level of the ovaries to the posterior end of the body (Figs 13–14). Sperm ducts form spermiducal vesicles laterally to the pharynx, diminishing in diameter laterally to the male copulatory apparatus (Fig. 15). They ascend, forming a loop, and then turn anteriad. Sperm ducts separately penetrate the penis bulb (Fig. 15) and open into the posteriorly directed diverticulum of the large bulbar cavity, in close proximity to each other (Fig. 16). Bulbar cavity single, ovoid, communicating with the ejaculatory duct through a short and narrow transition section (Figs 17–18). Ejaculatory duct opening at the tip of the conical and symmetrical penis papilla. The latter occupies almost the entire male atrium (Figs 13–15, 17–18).

Sperm ducts lined with a ciliated, cuboidal epithelium, underlain by a circular muscle layer. The large penis bulb consists of a loose connective tissue with weak interwoven muscle fibres (Figs 13–15, 17). Bulbar cavity lined with a ciliated, columnar epithelium, and coated with a subepithelial circular muscle layer, followed by a longitudinal muscle layer. Numerous glands with intrabulbar cell bodies and amorphous, cyanophil secretion open into the bulbar cavity. The bulbar cavity contains sperm and cyanophil secretion (Fig. 16). The transition part between the bulbar cavity and the ejaculatory duct is lined with a non-ciliated, cuboidal epithelium, the cells of which show an irregular height. A layer of circular muscle fibres surrounds the epithelial lining of the transition section. Ejaculatory duct lined with non-ciliated, cuboidal epithelium with irregular height, and surrounded by a subepithelial layer of circular muscle, followed by a layer of longitudinal muscle. This duct receives a xanthophil, fine granular secretion from intrapapillar glands. Penis papilla lined with a ciliated, columnar epithelium that becomes flat towards the tip of the papilla. Muscularis of penis papilla composed of a subepithelial layer of circular fibres and a layer of longitudinal fibres. Glands with amorphous, cyanophil secretion and with amorphous, xanthophil secretion open through the epithelial lining of the penis papilla. Both glands show intrapapillar cell bodies. Male atrium lined with a ciliated, high columnar epithelium, the cells of which have an irregular height (Fig. 17). The male atrial muscularis is constituted of a subepithelial layer of circular muscle, followed by a layer of longitudinal muscle. Glands with fine granular, cyanophil secretion and less numerous xanthophil glands with amorphous secretion open into the male atrium.

Female reproductive system (Figs 18, 19–22). Vitellaria poorly developed, located between intestinal branches. Ovaries, 70 μm – 100 μm in diameter, situated dorso-medially to the nerve cords, at about 0.6 mm from the anterior tip and close to the brain (110 μm posteriorly to the brain) (Fig. 19). Ovitelline ducts arising from the lateral surface of the ovaries (Fig. 20) and running backwards dorsally to the nerve cords. At about the level of the gonopore, the ovitelline ducts turn dorso-medially and form a common ovitelline duct that is dorso-anteriorly directed (Figs 18, 21). Common ovitelline duct opening into a female genital duct. The latter communicates with



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Figure 18. *Hausera hauseri* Leal-Zanchet & Souza, sp. n., holotype: sagittal composite reconstruction of the copulatory apparatus.

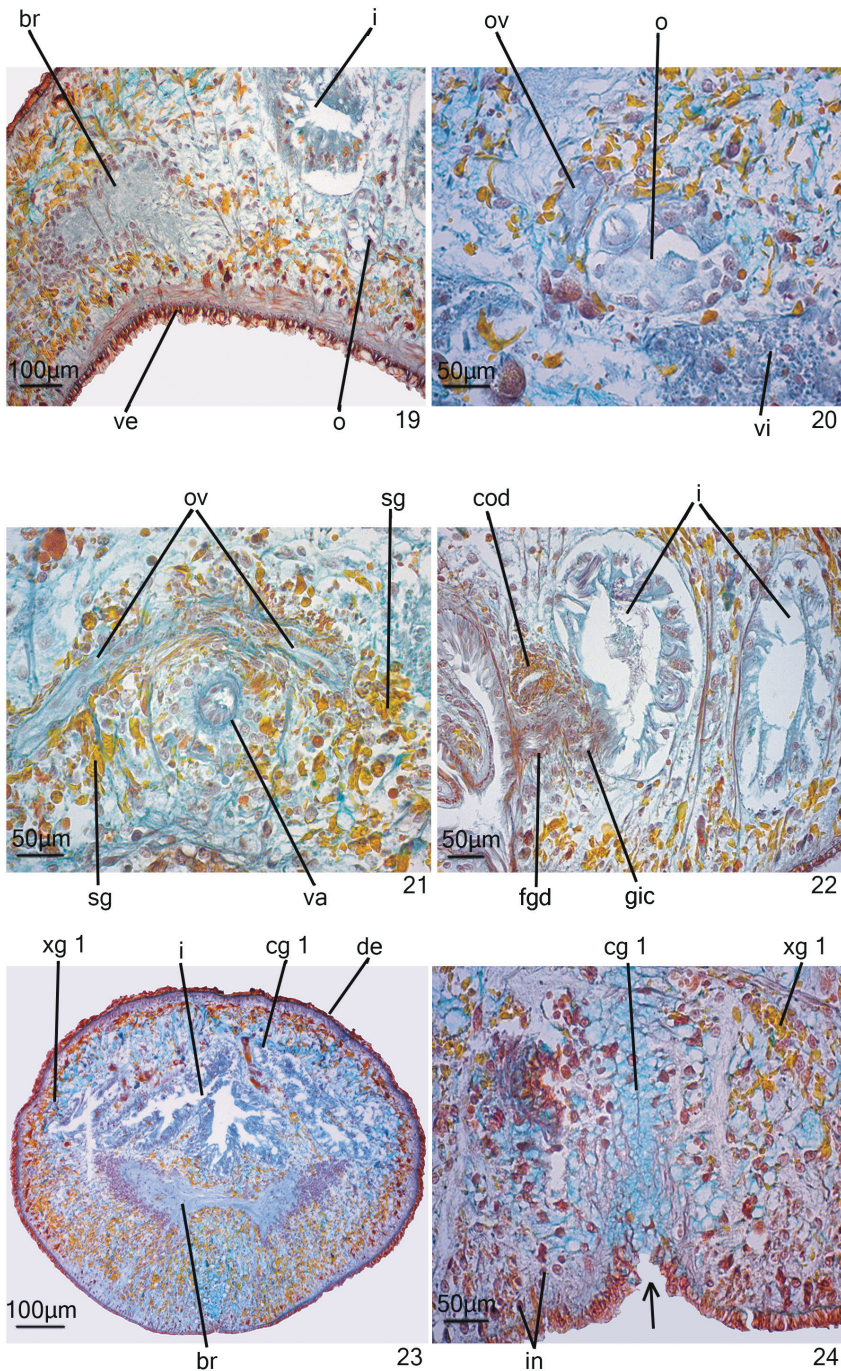
the male atrium and through a genito-intestinal canal with the intestine. The genito-intestinal canal is ventro-posteriorly directed (Figs 18, 22).

Ovovitelline ducts lined with ciliated, cuboidal epithelium, receiving the secretion of shell glands in their most posterior sections (Figs 18, 21). Common ovovitelline duct lined with ciliated, cuboidal or columnar epithelium, underlain by a circular muscle layer; it receives the secretion of shell glands (Fig. 18). These glands have a xanthophil, fine granular secretion (Fig. 21). Female genital duct and genito-intestinal canal lined with ciliated, cuboidal epithelium, receiving an amorphous, cyanophil secretion. The muscularis of the female genital duct and the genito-intestinal canal consists of a layer of circular fibres.

Etymology. The new species is dedicated to the late Prof Dr Josef Hauser SJ.

Geographical distribution. Known only from the type-locality, Felipe Guerra (Crotes cave), Brazil.

Variability. Vitellaria well-developed in paratype MZU PL.00148 and poorly developed in paratypes MZU PL.00142 and PL.00149. In paratype MZU PL.00148, the anterior region of the body is subcylindrical in transverse section (Fig. 23) and shows a ventral median slit, posteriorly to the brain, where abundant cyanophil secretion is discharged (Fig. 24). In this paratype, the anterior intestinal branch extends slightly anterior to the brain. A short branch of each ovovitelline duct may extend anterior to the ovaries at least in paratype MZU PL.00148.



Figures 19–24. *Hausera hauseri* Leal-Zanchet & Souza, sp. n.: holotype (19; 22) and paratype MZU PL.00148 (20–21; 23–24). 19 cephalic region in sagittal section 20 detail of the ovary and ovovitelline duct in transverse section 21–22 detail of the copulatory apparatus in transverse section (21) and sagittal section (22) 23–24 cephalic region in transverse section. The arrow indicates a slit in the ventral surface of the body.

Discussion

There are no genera within the Dimarcusidae into which we could comfortably include the species herein described; thus, the new genus *Hausera* is here proposed. Besides the combination of characters discussed above, the new genus is characterized by two unique features within its family: intestinal branch extending dorsally to the brain and ovovitelline ducts located dorsally to the nerve cords. According to Sluys (1990), the absence of a precerebral intestinal branch is a primitive character of the Dimarcusidae. Within this family, a precerebral intestinal branch is found only in *Opisthobursa josefinae* (Sluys 1990). In specimens of *H. hauseri*, the intestine generally extends dorsally to the brain, while it extends a little anterior to the brain only in paratype MZU PL.00148. The ovovitelline ducts are located medially to the nerve cords in the genera *Rhodax*, *Opisthobursa* and *Novomitchellia*, whereas they are located dorso-laterally to these cords in *Balliania* (Sluys 1990). The situation in *Hausera* differs from all these genera, in that the ovovitelline ducts are exactly dorsal to the nerve cords.

The type-locality of *H. hauseri*, a karst area located in Felipe Guerra, is unique in comparison with other karst areas in Brazil. Most limestone formations in Brazil are located in inner portions of the country, which in the past must have prevented marine groups from colonizing these caves. In contrast, the Felipe Guerra karst is located near the sea, and its limestone outcrops are at low altitude, which has allowed different invertebrates to colonize the caves during sea level rises in the past. Accordingly, many stygobiotic species found there represents oceanic relicts. They have evolved from marine ancestors trapped in the caves after isolation by events of introgression and regression of the ocean in the area, probably during the Tertiary Period. This is the case of stygobiotic amphipods (Ferreira et al. 2010, Fiser et al. 2013) and may apply also to *H. hauseri*.

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Revision of the paraphyletic genus *Koerneria* Meyl, 1960 and resurrection of two other genera of Diplogastridae (Nematoda)

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Abstract

Recent inferences of phylogeny from molecular characters, as well as a reexamination of morphological and biological characters, reject the monophyly of the nematode genus *Koerneria* Meyl, 1960 (Diplogastridae). Here, *Koerneria* sensu lato is revised. The genus, which previously consisted of 40 species, is separated into three genera. Almost all of the transferred species are moved to the resurrected genus *Allodiplogaster* Paramonov & Sobolev in Skrjabin et al. (1954). *Koerneria* and *Allodiplogaster* are distinguished from each other by a weakly vs. clearly striated body surface, an undivided vs. divided stomatal cheilostom, and arrangement of the terminal ventral triplet of male genital papillae, namely in that v5 and v6 are paired and separated from v7 vs. v5–v7 being close to each other. *Allodiplogaster* is further divided into two groups of species, herein called the *henrichae* and *striata* groups, based on both morphological and life-history traits. The *henrichae* group is characterized by papilliform labial sensilla and male genital papillae, a conical tail in both males and females, and an association with terrestrial habitats and insects, whereas the *striata* group is characterized by setiform labial sensilla and male genital papillae, an elongated conical tail in both sexes, and an association with aquatic habitats. A second genus, *Anchidiplogaster* Paramonov, 1952, is resurrected to include a single species that is characterized by its miniscule stoma and teeth, unreflexed testis, and a distinct lack of male genital papillae or stomatal apodemes. Lastly, one further species that was previously included in *Koerneria* sensu lato is transferred to the genus *Pristionchus* Kreis, 1932. The revision of *Koerneria* sensu lato is necessitated by the great variability in its subordinate taxa, which occupy a variety of habitats, in addition to the increased attention to Diplogastridae as a model system for comparative mechanistic biology.

Keywords

Allodiplogaster, *Anchidiplogaster*, *Koerneria*, *Pristionchus*, revision, phylogeny, taxonomy

Introduction

Koerneria Meyl, 1960 heretofore consisted of 40 nominal species, following the revision by Sudhaus and Furst von Lieven (2003) and including species described since then (Suppl. material 1). Several unidentified or undescribed species with molecular vouchers have also been reported (Mayer et al. 2007). Biological characters are variable for the genus, which contains terrestrial species isolated from rich soil environments, associates of several different groups of insects, and aquatic species. Such a diversity of ecologies raises the question of whether distinct subgroups of the genus can be identified and corroborated by independent characters.

In their revision of Diplogastridae, Sudhaus and Furst von Lieven (2003) circumscribed *Koerneria* by the following characters: (1) presence of stomatal dimorphism, (2) cheilostom often separated into six per- and interradial plates or small stick-like plates (= rugae), (3) vertical striation, (4) stegostom with a dorsal claw-like tooth, a right subventral tooth, and left subventral serrated plates, (5) postdental region of stegostom with two subventrad directed apodemes, (6) female gonad amphidelphic or seldom prodelphic, and (7) no bursa. Later, Furst von Lieven (2008) transferred *K. colobocerca* (Andrassy, 1964) Furst von Lieven, 2008 from *Mononchoides* Rahm, 1928 and revised the generic definition by adding two characters, (8) intestine sometimes with a prerectum, and (9) tail filiform or conical. Given the breadth of the most recent morphological definition of the genus, the only absolute generic character is the presence of articulated apodemes in the stoma. However, phylogenetic studies of molecular characters strongly indicate that the genus, as currently interpreted, is paraphyletic and thus separable into two or more clades (Fig. 1). Therefore, a taxonomic reorganization of *Koerneria* sensu lato (= *Koerneria* sensu Furst von Lieven 2008) is needed. Such a revision is of particular importance for ongoing studies of the natural history of the genus and its relatives (e.g., Giblin-Davis et al. 2006, Kanzaki et al. 2009) as well as comparative research more generally in Diplogastridae (e.g., Mayer et al. 2009, Ragsdale et al. 2013), as *Koerneria* sensu lato includes taxa that are sister groups to most other known species in the family.

In this article, we revise the genus *Koerneria* by examining the original and subsequent descriptions of its nominal species. Based on morphological, biological, and molecular evidence, we separate *Koerneria* sensu lato into three genera. All renamed groups are hypothesized to be monophyletic and follow the precedent of previous classification systems (Andrassy 1984). Besides limiting the scope of *Koerneria*, we resurrect the genera *Anchidiplogaster* Paramonov, 1952 and *Allodiplogaster* Paramonov & Sobolev in Skrjabin et al. (1954). Furthermore, we distinguish two putative clades of *Allodiplogaster* species, each of which we hypothesize to be monophyletic on available information, although we leave formal revision of this genus to follow molecular studies of unsampled aquatic taxa.

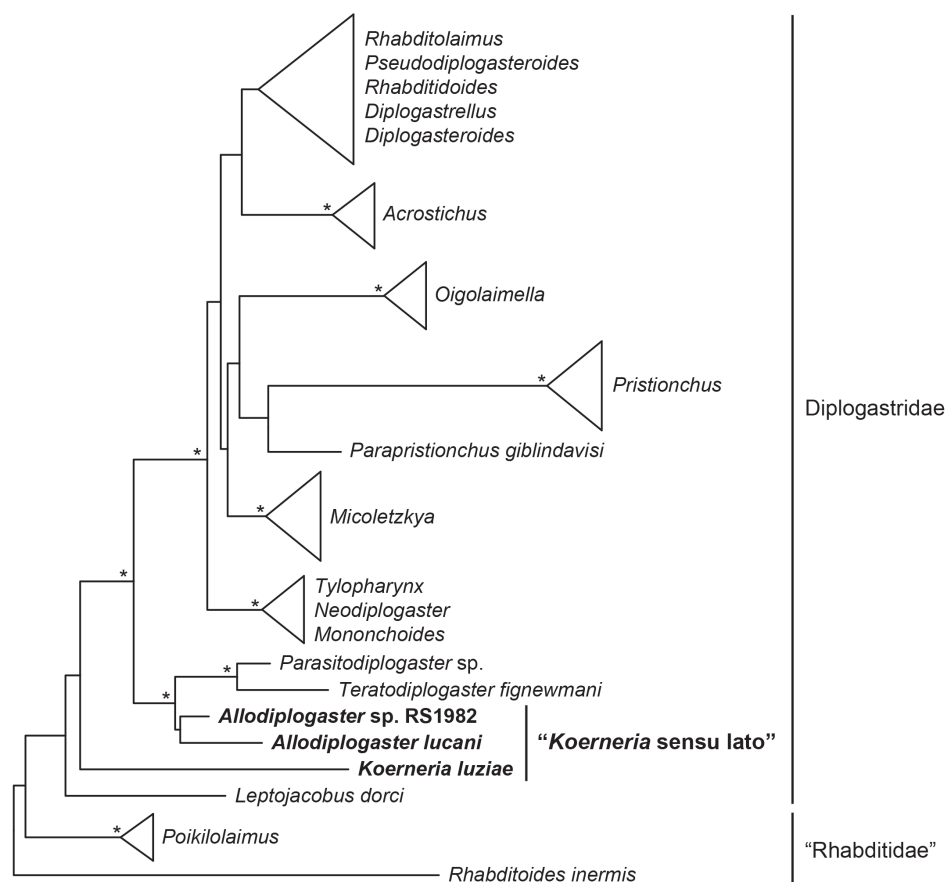


Figure 1. Paraphyly of *Koerneria sensu lato*. Tree is simplified from Kanzaki et al. (2014b), which was inferred from nearly full-length small subunit ribosomal DNA sequences. A subsequent study that included several more species of *Koerneria sensu lato* likewise showed the well-supported exclusion (99% bootstrap support in likelihood analysis, 100% posterior probability in Bayesian analysis) of a clade of *K. luziae* + *K. ruehmi* from all other Diplogastridae, including a monophyletic clade of what is designated herein as *Allodiplogaster* (Atighi et al. 2013). Asterisks indicate nodes with very strong support as inferred in the former study (>95% bootstrap support, 100% posterior probability).

Materials and methods

Species of *Koerneria sensu lato* are classified herein into four typological groups (three genera, with one genus further separated into two morphological and ecological groups) based on the following characters or traits, which were selected due to their high availability and reliability, being relatively accurate even in old descriptions:

- (1) Stomatal morphology, specifically the separation of cheilostom and presence of apodemes
- (2) Male tail morphology, including shape and arrangement of genital papillae

- (3) Female tail morphology
- (4) Life-history characters, particularly habitat preferences

In addition to published literature, several species available in culture were examined for typological characters: *Koerneria luziae*, isolated from stag beetles from Japan (Kanzaki et al. 2011); *Koerneria* sp., isolated from *Dorcus rectus* (Coleoptera: Lucanidae) from Japan; *Allodiplogaster* spp. RGD227 and RGD228, both isolated from soil-dwelling bees in the United States. Stomatal morphology and tail characters were examined by differential interference contrast (DIC) microscopy according to methods described by Kanzaki (2013). Several schematic illustrations were prepared based on original observations as well as published data. Following the reexamination of informative characters, the original descriptions and revisions were reviewed to determine the generic status of species following the International Code of Zoological Nomenclature (ICZN).

Systematics

Previous inferences of the phylogeny of *Koerneria* spp. indicate that two species, *K. luziae* (Körner, 1954) Meyl, 1960 and *K. ruehmi* Atighi, Pourjam, Kanzaki, Giblin-Davis, De Ley, Mundo-Ocampo & Pedram, 2013, form a well-supported clade that is the sister group to most or all other sequenced Diplogastridae (Atighi et al. 2013; Kanzaki et al. 2014b; Fig. 1). Separate from this group is a well-supported clade consisting of *K. lucani* (Körner, 1954) Meyl, 1960 and some unidentified or undescribed species (Fig. 1). The former two species share two features that clearly distinguish them from other species of *Koerneria* sensu lato, namely (1) a tube- or ring-like (undivided) cheilostom and (2) male genital papillae arranged such that v5 and v6 (papillae nomenclature follows Sudhaus and Fürst von Lieven 2003) are close to each other and v7 is clearly apart from v6. These two characters diagnose six nominal species of *Koerneria* sensu lato, including the above two as well as the type species of the genus. We therefore revise the genus as follows:

Koerneria Meyl, 1960

Fig. 2

Generic diagnosis

- 1) Stomatal dimorphism occasionally present¹
- 2) Body-wall cuticle with weak vertical striations
- 3) Cheilostom usually forming short, undivided tube; rugae absent
- 4) Stegostom with dorsal claw-like tooth, right subventral tooth, and left subventral serrated plates or ridges

¹ Dimorphism has been confirmed in at least two species in this group, including one undescribed species (Kanzaki and Ragsdale et al. unpubl. obs.)

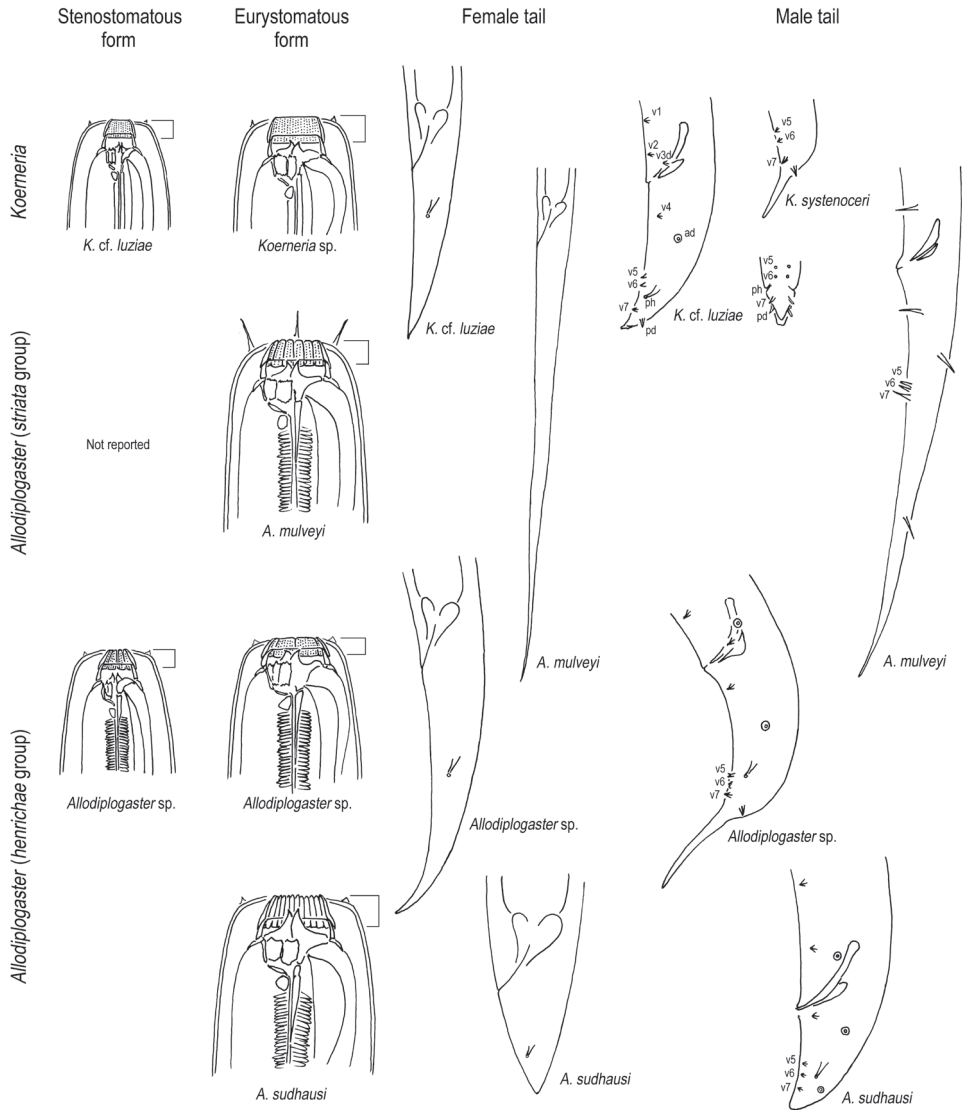


Figure 2. Schematic drawings of the generic characters of *Koerneria* and *Allodiplogaster*. From left to right: stenostomatous form, eurystomatous form, female tail, and male tail characters. From top to bottom: *Koerneria*, aquatic *Allodiplogaster* (“*striata*” species group) and two types of terrestrial *Allodiplogaster* (“*henrichae*” species group). For the stenostomatous form of *A. sudhausi* (not shown), see Fürst von Lieven (2008). Squared bracket indicates cheilostom, which as undivided separates *Koerneria* from *Allodiplogaster*. Further diagnosing *Koerneria* is the arrangement of male genital papillae v5-v7. Unique to the *striata* group of *Allodiplogaster* relative to the *henrichae* group and to *Koerneria* are a long tail in both sexes, setiform genital papillae, and in many cases setiform labial papillae. Genital papillae and phasmids are labeled following the terminology in Sudhaus and Fürst von Lieven (2003). The phasmids are not clearly described in species of the *striata* group.

- 5) Postdental region of stegostom with left and right apodemes directed subventrad
- 6) Female gonad amphidelphic
- 7) Anterior two ventral and distal pairs of genital papillae (v5 and v6) close to each other, the posterior pair (v7) being clearly apart from v6
- 8) Short and conical male tail (c' is usually ≤ 3) usually with short spike or with small, bursa-like or membranous appendage at tail tip
- 9) Known from terrestrial habitats, often in association with insects

Type species

Diplogaster goffarti Körner, 1954

comb. *Koerneria goffarti* Meyl, 1960

Other species

K. erlangensis (Sachs, 1950) Sudhaus & Fürst von Lieven, 2003

K. luziae (Körner, 1954) Meyl, 1960

K. ruehmi Atighi, Pourjam, Kanzaki, Giblin-Davis, De Ley, Mundo-Ocampo & Pedram, 2013

K. sinodendroni (Körner, 1954) Meyl, 1960

K. systemoceri (Körner, 1954) Meyl, 1960

Following this restricted definition of *Koerneria*, most of the remaining species of *Koerneria* sensu lato are transferred to the resurrected genera *Anchidiplogaster* and *Allodiplogaster*. One species, which was previously combined as *K. dubia* (Hnatewytch, 1929) Sudhaus & Fürst von Lieven, 2003 is returned to *Anchidiplogaster* based on a suite of characters unique to this taxon as well as by the lack of stomatal apodemes, an absolute character of *Koerneria* as defined both previously and herein:

Anchidiplogaster Paramonov, 1952

Generic diagnosis

- 1) Miniscule, undivided stoma with two small, similarly sized pyramidal teeth (one dorsal and one right subventral)
- 2) Stomatal apodemes absent
- 3) Male genital papillae absent
- 4) Testis without flexure

Type and only species

=*Diplogaster dubia* Hnatewytch, 1929

comb. *Anchidiplogaster* Paramonov, 1952

All but one of the remaining species of *Koerneria* sensu lato are transferred to the other resurrected genus, *Allodiplogaster*. This name has priority (ICZN 23.1) over other names that are available for this taxonomic grouping, which consist of *Diplenteron* Andrassy, 1964, *Glauxinemella* Gagarin, 1998, and *Gobindonema* Khera, 1970. Furthermore, *Al-*

lodiplogaster is separated into two putatively monophyletic groups of species, which we designate as the “*henrichae* group” and “*striata* group” based on morphological and biological evidence.

***Allodiplogaster* Paramonov & Sobolev in Skrjabin, Shikobalova, Sobolev, Paramonov & Sudarikov, 1954**

Fig. 2

=*Diplenteron* Andrásy, 1964: *D. colobocercus* Andrásy, 1964

=*Gobindonema* Khera, 1970: *G. filicaudata* Khera, 1970

nec *Gobindonema* Sood & Prashad, 1974 (Trichostrongylidae)

=*Glauxinemella* Gagarin, 1998: *G. striata* Gagarin, 1998

Generic diagnosis

- 1) Stomatal dimorphism occasionally present
- 2) Body-wall cuticle with clear vertical striations
- 3) Cheilostom separated into six per- and interrational plates or rugae
- 4) Stegostom with dorsal claw-like tooth, right subventral tooth, and left subventral serrated plates or ridges
- 5) Postdental region of stegostom with left and right apodemes directed subventrad
- 6) Female gonad amphidelphic; rarely prodelphic
- 7) Distal triplet papillae of males (v5-7) close to each other
- 8) Tail of male and females highly variable in shape
- 9) Known from variable habitats including terrestrial insect associates and aquatic species

Type species

Diplogaster henrichae Sachs, 1950

comb. *Allodiplogaster henrichae* Paramonov & Sobolev in Skrjabin, Shikobalova, Sobolev, Paramonov & Sudarikov, 1954

Other species

henrichae* group of *Allodiplogaster

- 1) Labial sensilla usually papilliform
- 2) Female tail usually conical with or without filiform tip
- 3) Male tail usually conical with short spike
- 4) Male genital papillae short and papilliform
- 5) Known from terrestrial habitats, often in association with insects

A. colobocerca (Andrásy, 1964), comb. n.

=*Mononchoides potohikus* Yeates, 1969

A. hirschmannae (Sachs, 1950), comb. n.

A. histophora (Weingärtner, 1955), comb. n.

A. hylobii (Fuchs, 1915), comb. n.

A. incurva (Körner, 1954), comb. n.

- A. labiomorpha* (Kühne, 1995), comb. n.
A. lepida (Andrássy, 1958), comb. n.
A. lucani (Körner, 1954), comb. n.
A. pierci (Massey, 1967), comb. n.
A. pini (Fuchs, 1931), comb. n.
A. robinicola (Rühm, 1956), comb. n.
A. sudhausi (Fürst von Lieven, 2008), comb. n.

striata* group of *Allodiplogaster

- 1) Labial sensilla setiform
- 2) Male and female tail usually elongate-conical, with or without filiform tip
- 3) Male genital papillae setiform
- 4) Known from aquatic habitats

- A. angarensis* (Gagarin, 1983), comb. n.
A. aquatica (Dassonville & Heyns, 1984), comb. n.
A. baicalensis (Tsalolichin, 1972), comb. n.
A. carinata (Zullini, 1981), comb. n.
A. didentata (Hnatewytch, 1929), comb. n.
 =*Diplogaster curvidentatus* Altherr, 1938
 =*Diplogaster obscuricola* Altherr, 1938
 =*Diplogaster quadridentatus* Altherr, 1938
A. filicaudata (Khera, 1970), comb. n.
A. ivanegae (Gagarin, 1983), comb. n.
A. lupata (Shoshin, 1989), comb. n.
A. mordax (Shoshin, 1989), comb. n.
A. mulveyi (Ebsary, 1986), comb. n.
A. pantolaba (Shoshin, 1989), comb. n.
A. pararmata (Schneider, 1938), comb. n.
 =*Diplogaster armatus apud* Filipjev, 1930, *nec* Hofmänner, 1913
A. regia (Shoshin, 1989), comb. n.
A. ruricola (Gagarin, 1983), comb. n.
A. sphagni (Soós, 1938), comb. n.
A. strenua (Gagarin, 1983), comb. n.
A. striata (Gagarin, 1998), comb. n.
A. tenuipunctata (Altherr, 1938), comb. n.
A. terranova (Ebsary, 1986), comb. n.

The single remaining species previously included in *Koerneria* sensu lato is transferred to *Pristionchus* Kreis, 1932. This transfer is supported by the absence of subventral apodemes, the key character diagnosing *Koerneria* sensu lato:

- Pristionchus macrospiculum* (Altherr, 1938), comb. n.

Discussion

The separation of *Koerneria* as circumscribed herein from *Allodiplogaster*, *Anchidiplogaster*, and *Pristionchus* is strongly supported by the structure of the cheilostom, arrangement of male genital papillae, and phylogeny as inferred independently from molecular sequence characters (Atighi et al. 2013; Kanzaki et al. 2011, 2013, 2014b). Although for the previous, wider definition of *Koerneria* the name *Koerneria* was itself a junior synonym of *Allodiplogaster*, the name *Koerneria* is retained for a group of six described species, all of which are unambiguously unified by morphological characters and which are represented by a clade not nested within any other valid genus. Furthermore, species of *Koerneria* in the revised sense are apparently the sister group to most or all species of Diplogastridae, with the possible one exception of *Leptojacobus dorci* (see Kanzaki et al. 2014b), and therefore the revision of this genus will be useful for ongoing research on the family as a comparative model system.

The distinct morphology of *Anchidiplogaster dubia*, especially its lack of genital papillae in several observed male specimens separates this species from all other Diplogastridae and supports its reestablishment in that genus. Further indicating the distinctness of this species from most other known Diplogastridae is its unreflexed testis flexure, as a flexure was previously considered a plesiomorphic character of the entire family (Sudhaus and Fürst von Lieven 2003, Andr  ssy 2005), although one other diplogastrid genus, *Leptojacobus* Kanzaki, Ragsdale, Susoy & Sommer, 2014, has since been described to have no flexure. In the original description of *A. dubia*, the above missing features were explicitly given as diagnostic characters (Hnatewytsh 1929) and thus were unlikely to be simply missed in all specimens examined. An undivided stoma is also unusual among most diplogastrids with subventral teeth, although this type of stoma is present in *Koerneria* in the revised sense and is thus insufficient in itself to diagnose *Anchidiplogaster*. The presence of asymmetrical teeth and a glandular postcorpus clearly support its identity as a diplogastrid, the otherwise divergent stomatal morphology in this species, namely its diminutive, undivided stomatal cavity armed with triangular teeth, obscure its relationships to other taxa within the family. Because molecular characters are not available for this species, its position in Diplogastridae cannot be predicted with any certainty.

The split of *Allodiplogaster* into the *henrichae* and *striata* groups is also supported by morphology, principally by the tail of both sexes, which is usually much longer in *striata* group than in *henrichae* group, and by the male genital papillae and labial sensilla, which are distinctly setiform in *striata* group. The separation of the two proposed groups of *Allodiplogaster* is only confounded by overlapping morphological characters, namely the fish-bone-like swellings along the pharyngeal lumen that are present in *henrichae* group species, *Allodiplogaster* spp. RGD227 and RGD228, as well as in one species of the *striata* group, *A. carinata*, although not in another *striata* group species such as *A. pararmata* (F  rst von Lieven 2001, F  rst von Lieven and Sudhaus 2000, Kanzaki et al. unpubl. obs.). However, no species of the *striata* group have been molecularly characterized thus far, so the phylogenetic position of that group has yet to be tested

by gene sequence data. Reisolation of species, particularly of the *striata* group, will be necessary for fully testing the system of intrageneric grouping presented here. Complete taxonomic revision of *Allodiplogaster* is therefore not performed here due to a lack of molecular evidence, although further studies may confirm the separation of the two species groups designated here into different genera.

Other morphological characters may further support to separate *henrichae* group from as a monophyletic clade distinct from the *striata* group and from *Koerneria*. Kühne (1995) examined three *henrichae* group species by scanning electron microscopy and reported important morphological features of the male tail: the modification in v5 and v6 subventral distal papillae, whereby v5 is rooted in a socket-like base and has split tip, and v6 having two anterior and posterior appendages and being likewise rooted in a socket-like base (Fig. 2). This morphology, which is similar to the trifurcate genital papillae of *Diplogasteroides* spp. (Kiontke et al. 2001, Kanzaki et al. 2013), is otherwise unique among Diplogastridae and Rhabditidae thus far characterized (e.g., Kiontke and Sudhaus 2000, Kanzaki et al. 2012a), and has additionally been found in two undescribed *henrichae* group species (Giblin-Davis et al. unpubl. obs.). The diagnostic utility of modified genital papillae has already been shown for another clade of Diplogastridae: in *Pristionchus* and the closely related genus *Parapristionchus* Kanzaki, Ragsdale, Herrmann, Mayer, Tanaka & Sommer, 2012, the v5 and v6 papillae are shrouded at the base by a socket-like structure, where the tip of v6 is split into two papilla-like projections (e.g., Kanzaki et al. 2012b, 2014a). Therefore, further examination of male tail morphology in other species of *Koerneria* sensu lato may confirm the importance of v5 and v6 as diagnostic of the *henrichae* group of *Allodiplogaster*. Yet another putative character for the *henrichae* group is the presence of a prerectum, as suggested to distinguish the “*Diplenteron* group” of *Koerneria* sensu lato (Fürst von Lieven 2008) and which was reported for *A. colobocerca* and *A. sudhausi*. The prerectum, which is a shallow but distinct constriction separating the posterior part of the intestine from the anterior part, can be found in three undescribed *Allodiplogaster* species (Kanzaki and Giblin-Davis unpubl. obs.), lending preliminary support to this idea. This character is not present in *Koerneria* nor in any species of *striata* group as interpreted from original descriptions, and therefore it may additionally support monophyly of the *henrichae* group.

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Supplementary material I

Species status and several key characters of *Koerneria sensu lato* in the revised genera.

Authors: Natsumi Kanzaki, Erik J. Ragsdale, Robin M. Giblin-Davis

Data type: Summary of typological characters as a spread sheet.

Explanation note: Taxonomic history, morphological (typological) characters, and life-history traits of *Koerneria sensu lato* in the revised genera.

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A new genus of Strepsiptera, *Rozenia* gen. n. (Stylopidae), a parasite of bee genera *Acamptopoeum* and *Calliopsis* (Andrenidae, Panurginae, Calliopsini)

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Abstract

A new Strepsiptera genus from South America is described, *Rozenia* gen. n., with three new species: *R. calliopsidis* sp. n. (type species), *R. peruana* sp. n. and *R. platicephala* sp. n. These three new species are parasites of bees belonging to the tribe Calliopsini (Andrenidae, Panurginae). *Rozenia calliopsidis* sp. n. is a parasite of the bee genus *Calliopsis* Smith, 1853 and *R. peruana* sp. n. and *R. platicephala* sp. n. are parasites of the bee genus *Acamptopoeum* Cockerell, 1905. Diagnoses and descriptions of female puparia are presented for all three species. Diagnoses and descriptions of first instars (triungulinids) are presented for *R. calliopsidis* sp. n. and *R. platicephala* sp. n. The first case of increased number of setae on the body of the first instars and augmentation of chaetotaxy of Strepsiptera are discussed.

Keywords

New genus, new species, Apoidea, host-parasite association, morphology, chaetotaxy, description, South America

Introduction

Stylopization of a bee from the tribe Calliopsini (Andrenidae, Panurginae) was recorded for the first time as early as 1931 (Schwarz 1931). Another finding was presented shortly afterwards from Argentina (Hofeneder and Fulmek 1943). Both records were repeated in the literature several times under various combinations of the host names (Hofeneder and Fulmek 1943, Hofeneder 1949, 1952) that recently belong to the genus *Acamptopoeum* Cockerell, 1905. However, no other record of stylopization of a bee from the tribe Calliopsini has been published since that time.

Members of the tribe Calliopsini are not the only known stylopized panurgine bees. Pierce (1904) published a note on a stylopized bee from the tribe Protandrenini and subsequently added data on stylopization of a wider range of species from the tribe Protandrenini from North America (Pierce 1909). All these North American panurgine hosts of Strepsiptera belong to the genus *Pseudopanurgus* Cockerell, 1897; Strepsiptera parasitizing Protandrenini are also known from South America (Holmberg 1921, Ogloblin 1947, Kogan 1989). These hosts belong to the genera *Anthrenoides* Ducke, 1907, *Psaenythia* Gerstaecker, 1868 and *Rhophitulus* Ducke, 1907. There are also two other genera of Panurgini known to be hosts of Strepsiptera in the Palearctic region. The first note about stylopized *Panurgus* Panzer, 1806 (Panurgini) was made by Morice (1913) and later, Ogloblin (1925) recorded Strepsiptera from the genus *Panurginus* Nylander, 1848.

All described Strepsiptera, which parasitize panurgine bees, were placed in the genus *Crawfordia* Pierce, 1908. All bees were from Neotropical, Nearctic, or Palearctic regions. Regarding the Strepsiptera that parasitize bees from the tribe Calliopsini, no taxon has ever been described, even though the host-parasite association has been known for more than eighty years. Here we present a new genus of Strepsiptera associated with the bee tribe Calliopsini, with a description of three new species. We compare the morphology of female puparia and the first instars with other genera of Strepsiptera, and particularly with species parasitizing bees (Stylopidae), especially other members of the bee subfamily Panurginae.

Methods

Collections

Material from the following public and private collections was examined:

- AMNH** American Museum of Natural History, Jerome G. Rozen Jr., (New York, USA);
- JSPC** Jakub Straka personal collection, (Praha, Czech Republic);
- KUNHM** Natural History Museum, Division of Entomology, University of Kansas, Michael S. Engel, (Lawrence, Kansas, USA).

Preparation of material

All host individuals were first relaxed and then dissected. Females and first instar larvae were removed from the host body. Strepsiptera females studied for morphology were cleared using proteinase: a mixture of lysis buffer and proteinase K (Quiagen) was heated to 56 °C. The lysis procedure took several hours or overnight. Cleared specimens were cleaned in water several times and then stored in vials with glycerol. A drawing tube (camera lucida) was attached to an Olympus BX40 light microscope and an Olympus SZX9 binocular microscope and used for morphological studies and drawings. Temporary slides were prepared with glycerol.

First instar larvae were removed from the female's body. Specimens used for morphological studies were prepared using the same method as females, except for scanning electron microscopy (SEM). For SEM, first instars were stored in 96% ethanol and subsequently dehydrated in 100% ethanol for 5–10 minutes and then acetone for 5 minutes. Dehydrated specimens were critical-point dried and coated with gold. For scanning electron microscopy we used a JEOL 6380 LV.

Morphology and terminology

External structures of first instars and female puparia are described. The mature or tenereal female is presented inside the external puparium, but these have never been used for species descriptions. The body is weakly sclerotized and, in addition to the number of birth organs (tubae proliferae), lacks any practical characters.

Morphological terminology of female puparium follows Kinzelbach (1971) except:

basal band	pigmented external part of abdominal segment I, usually distinct on ventral side;
cephalic ridge	intersegmental ridge between head and prothorax on ventral side;
cintum	constriction dividing inner and outer part of tergum I;
head corner	lateral extensions of head behind brood opening on ventral side;
oral ridge	mouth sensu Kinzelbach (1971);
prothoracic ridge	intersegmental ridge between prothorax and mesothorax on ventral side.

Terminology of first instar larvae follows Pohl (2000, 2002) except:

interstitial row of setae	additional row of setae between submedian and supralateral row on thoracic tergites.
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Specimens of strepsipterans are indicated by the following abbreviations: EMP – empty male puparium; MP – male puparium; FP – female puparium; L1 – first instar larva.

Description style

All newly described species were labeled as follows: “HOLOTYPUS FP, name of taxon sp. nov., Jakub Straka det. 2014” on red card; paratypes analogously on yellow card. Precise label data on locality are cited for the holotypes. Separate lines on a label are indicated by a slash “/” and separate card labels are indicated by a double slash “//”.

Information on the distribution and etymology of names are provided in separate paragraphs for each species. An overview of the host-parasite associations with published and updated host names is presented in Table 1. Information concerning host stylopization without classification of the Strepsiptera and other information are within the notes.

Table 1. Summary of host associations for *Rozenia* gen. n. All hosts belong to bees (Apoidea) of the family Andrenidae; (as) host published under the different combination or misidentification; (*) host association corroborated in this study. Valid names are in bold.

Parasite	Host
Strepsiptera	Hymenoptera
Stylopidae	Panurginae Leach, 1805 (Apoidea: Andrenidae Latreille, 1802)
<i>Rozenia</i> gen. n.	Calliopsini Robertson, 1922
<i>R. calliopsidis</i> sp. n.	* <i>Calliopsis (Liopoeum) mendocina</i> (Jørgensen, 1912)
	* <i>Calliopsis (Liopoeum) trifasciata</i> (Spinola, 1851)
<i>R. peruana</i> sp. n.	* <i>Acamptopoeum vagans</i> (Cockerell, 1926)
<i>R. platicephala</i> sp. n.	* <i>Acamptopoeum submetallicum</i> (Spinola, 1851)
	as <i>Liopoeum submetallicum</i> (Spinola, 1851) (Schwarz 1931, Hofeneder and Fulmek 1943)
<i>Rozenia</i> sp.	<i>Acamptopoeum argentinum</i> (Fries, 1906)
	as <i>Perdita argentina</i> Fries, 1906 (Hofeneder and Fulmek 1943)
	as <i>Calliopsis (Parafriesea) argentina</i> (Fries, 1906) (Hofeneder 1952)

Genus and species descriptions

Rozenia gen. n.

<http://zoobank.org/00957F90-4A0F-4ACB-AAB5-9CFF8B9A303A>

Type species. *Rozenia calliopsidis* sp. n.

Diagnosis. Female. *Rozenia* gen. n. differs from other genera of the family Stylopidae in having only four abdominal segments. Similarly to the genus *Crawfordia* Pierce, 1908, canalis prolifer of *Rozenia* gen. n. is with a single median tuba prolifera present on segments II-IV of the abdominal part of female. However, tuba prolifera III of *Rozenia* gen. n. is positioned on the posterior half of abdominal segment IV, but in the middle of segment IV in *Crawfordia*, which possesses also rudimentary segment V. Abdomen of other genera of the family Stylopidae is composed by higher number of segments.

Female puparium. Brood opening of the new genus is very wide, almost from side to side, about four times wider than intermandibular distance, or more in *Rozenia* gen. n. Brood opening is usually much narrower in other genera of the family Stylopidae. Narrow head corners are produced laterally beyond prothorax; this feature causes head to be wider than distal part of prothorax and side of cephalothorax is not continuously diverging posteriorly. This character is developed in *Eurystylops* Bohart, 1943 and some species of the genus *Crawfordia*. Head corners are relatively long, but not as long as in *Crawfordia*, which possess head corners longer than half of cephalothorax. In *Rozenia* gen. n. head corners are as long as head dorsally, but *Crawfordia* has much longer head corners than head dorsally. Mandibles extending from the head contour in ventral view. In contrast to *Crawfordia*, intersegmental ridges are not developed in *Rozenia* gen. n.

First instar. First instars of *Rozenia* gen. n. differ substantially from other genera by having setae of submedian row on thorax as well as on abdominal segments. Caudal setae are distinctly longer than body. Both these characters are unique among all Strepsiptera. *Rozenia* gen. n. does not have spinulae on posterior margin of thoracic tergites as in Xenidae, Halictophagidae, or Elenchidae. These spinulae are developed on posterior margin of thoracic tergites in all other genera of the family Stylopidae. Ventral sublateral bristle is missing on sternum IX in *Rozenia* gen. n., but probably present in all other genera of Strepsiptera. Posterior margin of labiomaxillary area continuous in *Rozenia* gen. n., but emarginated in *Crawfordia*, *Halictoxenos* and *Stylops* (and probably also in other Stylopidae).

Description. Female. Canalis prolifer on abdominal segments I-IV, segment V absent. Single median tuba prolifera on segments II-IV, tuba prolifera on segment IV positioned in posterior half of segment.

Female puparium. Head corners (on ventral side) extending posteriorly as far as head posterior margin on dorsal side; head corners distinct, narrow, forming a lamella on frontal part of cephalothorax, produced laterally beyond prothorax, this feature causes head to be wider than distal part of prothorax and side of cephalothorax is not continuously diverging posteriorly; head corners elevated ventrally over intermandibular part of head, but not over prothorax; brood opening wide, distinctly wider than distance between mandibles; mandibles variable in size, but at least the tip is extending from the head contour in ventral view. Intersegmental ridges not developed; anterior margin of mesothorax ill-defined, but transverse and does not extend forward; spiracles positioned distally above prominent spiracular corners, close to middle of cephalothorax. Prothorax ventrally pigmented, not lighter than head corners.

Male. Unknown.

First instar. Body rounded; thorax approx. half of entire body length (caudal setae not included); caudal setae distinctly longer than body length. Head strongly reduced ventrally; maxilla with single seta; mandibles and labrum overlapping outline of body; labium fused to maxillae forming labiomaxillary area, its posterior margin continuous, not emarginated.

Each segment of thorax bears at least two pairs of setae dorsally and laterally close to posterior margin, forming submedian and lateral rows of setae. Posterior margins

of thoracic tergites smooth. Coxae broad, ovate; three coxal teeth at anterior part of each coxa, all divided into two to four tips; one coxal bristle divided at least into two tips; up to five cuticular outgrowths laterally from coxal teeth and one very short seta anteriorly from cuticular outgrowths; one very short seta at posterior part of coxa. Each trochanterofemur with femoral spur bifid at tip; up to six cuticular outgrowths and one short seta anteriorly and posteriorly on femur; each tibia with five tibial spurs and small projections at distal end of tibia. Tarsi of fore and mid legs enlarged and elongated; tarsi of hind legs rod-like and elongated. Sternal plates broad and smooth on surface (paired setae missing).

Abdomen with rows of setae similar to those present on thorax. Abdominal segment X extremely shortened and fused to segment IX; segment XI split into two parts and restricted to ventral base of caudal setae; segment XI with one particularly long caudal seta and short lateral caudal seta. Posterior margins of abdominal tergites smooth except laterally, spinulae not immersed; posterior margin of abdominal sternites with spinulae, spinulae not immersed; segment IX with only two spinulae, ventral sublateral bristle is missing.

Hosts. Bees of the genera *Acamptopoeum* and *Calliopsis*.

Etymology. Named in honor of the excellent bee expert, teacher of generations of bee students and a friendly and knowledgeable man, Jerome G. Rozen Jr. (American Museum of Natural History, New York, USA). J.G. Rozen Jr., collected most of the specimens of all three new species used for the descriptions.

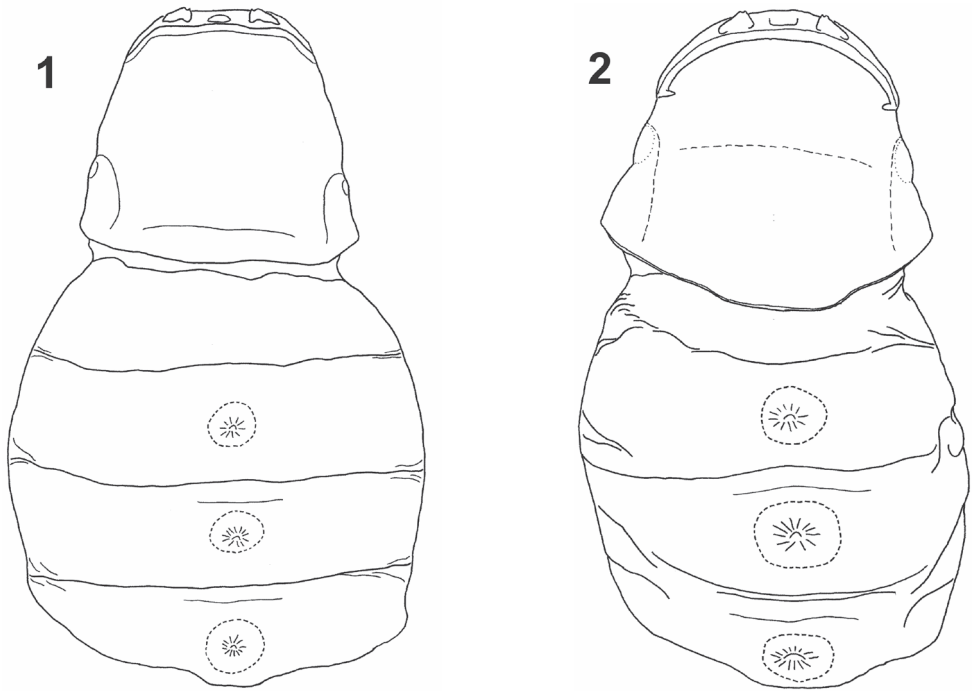
***Rozenia calliopsidis* sp. n.**

<http://zoobank.org/DABEEEE4-1BEF-4764-812D-054FDA6D8FD3>

Figures 1, 3, 6, 9, 12, 14, 17, 19, 20, 22

Material examined. Holotype female puparium, in a separate microvial on the same pin as host. Original label: “CHILE: R.M.: Chacabuco / Caleu, nr. Cerro del Robie / 33°00'49"S, 70°58'59"W / 30 Nov 2004, J. S. Ascher, / A. Y. Kawahara, C. Espina”. 1 FP, host: *Calliopsis* (*Liopoeum*) *trifasciata* (Spinola, 1851), ♂, AMNH coll. (code: AMNH_BEE 00036534).

Paratypes: ARGENTINA: Salta prov.: Cafayete, 14.xi.1993, 1 FP, host: *Calliopsis* (*Liopoeum*) *mendocina* (Jørgensen, 1912) ♀, JG and BL Rozen leg., AMNH coll. (AMNH_BEE 00036520), ditto, 1 FP (AMNH_BEE 00036521), ditto (AMNH_BEE 00036522); Catamarca prov.: El Desmonte, 7.xi.1989, 1 FP, host: *C. m.* ♂, JG Rozen and A Roig-Alsina leg., AMNH coll. (AMNH_BEE 00036523), ditto 2 FPP (AMNH_BEE 00036524), San Fernando, 3.–6.xi.1989, 1 FP, host: *C. m.* ♀, JG Rozen and A Roig-Alsina leg., AMNH coll. (AMNH_BEE 00036525), ditto, 5.xi.1991, 1 FP, host: *C. m.* ♂, JG Rozen, LE Peña and A Ugarte leg., AMNH coll. (AMNH_BEE 00036529), ditto, 15.xi.1993, 1 FP, host: *C. m.* ♂, JG and BL Rozen leg., AMNH coll. (AMNH_BEE 00036528), Tinogasta 35 km SE, 28.xi.1989, 1 FP, host: *C. m.* ♂, JG Rozen and A Roig-Alsina leg., AMNH coll. (AMNH_BEE 00036526), Co-



Figures 1–2. Female puparium, cephalothorax, with canalis prolifer of female, ventral view. **1** *Rozenia calliopsidis* sp. n. **2** *Rozenia platicephala* sp. n.

pacabana, 30.xi.1993, 1 FP, host: *C. m.* ♂, JG Rozen leg., AMNH coll. (AMNH_BEE 00036527), Punta de Balasto 3–15 km WSW, 25.xi.1993, 1 FP, host: *C. m.* ♀, JG Rozen leg., AMNH coll. (AMNH_BEE 00036530); Tucumán prov.: Amaichá del Valle, 6.iii.1990, 1 MP with pupa, host: *C. m.* ♂, JG Rozen leg., AMNH coll. (AMNH_BEE 00036532); Rio Negro prov.: El Bolson, 17.ii.1960, 1 FP, >50 L1, host: *C. t.* ♂, A Kovacs leg., AMNH coll. (AMNH_BEE 00036533); Neuquén prov.: Junín de los Andes, 21.–23.ii.2004, 2 FPP, host: *C. t.* ♀, J Straka leg. and det., JSPC coll.; CHILE: Apoquindo, Santiago, 1FP, host: *C. t.* ♂, date and collector not indicated, KUNHM coll. (SEMC1008235); Macul, SE Santiago, 5.xi.1974, 2 FPP, host: *C. t.* ♀, LE Peña leg., AMNH coll. (AMNH_BEE 00036536); Petorca prov.: Las Palmas tunnel, 18.x.1994, 2 FPP, host: *C. t.* ♂, JG Rozen, Quinter and JS Ascher leg., AMNH coll. (AMNH_BEE 00036535). Other material examined: Salta prov.: El Carmen, 27 km S Molinos, 1900 m, 6.x.1968, 1 EMP, host: *C. m.* ♀, LE Peña leg., AMNH coll. (AMNH_BEE 00036519). If not indicated otherwise, bee hosts identified by JS Ascher.

Diagnosis. Female puparium. *Rozenia calliopsidis* sp. n. differs from other species of the genus by a narrower head with large mandibles. Brood opening turned backwards laterally, very close to posterior margin of mandible and continued as cephalic ridge. In other species, the brood opening fluently transforms into cephalic ridge and forms an arcuate line. Spiracular corners of this species are weakly prominent, obtuse,

not triangular. Whole cephalothorax is darker than in *R. platicephala* sp. n. and *R. peruana* sp. n.

First instar. Shape of body narrower than in *R. platicephala* sp. n. Ratio of body length and width is on average 2.3. Ratio of body length and length of caudal setae is 0.74–0.96. Caudal setae are shorter than in *R. platicephala* sp. n.

Head dorsally with seven pairs of setae compared to six and usually shorter in *R. platicephala* sp. n. Labrum is not emarginated in the middle in contrary to *R. platicephala* sp. n. Labiomaxillary area more rounded than in *R. platicephala* sp. n., acute posteriorly.

Each segment of thorax bears only two pairs of setae dorsally and laterally, forming submedian and lateral row of setae, both rows continue on abdomen, interstitial and supralateral rows of setae missing. Posterior margin of abdominal tergites with more spinulae laterally than in *R. platicephala* sp. n. These spinulae are visible in dorsal view.

Sternal plates are broad and smooth on surface, posterior margin with fringe of long spinulae in contrast to smooth margins of *R. platicephala* sp. n. Precoxal pleural membrane of prothorax covered with transverse row of microtrichiae and precoxal pleural membrane of meso and metathorax with two cuticular processes laterally and medially.

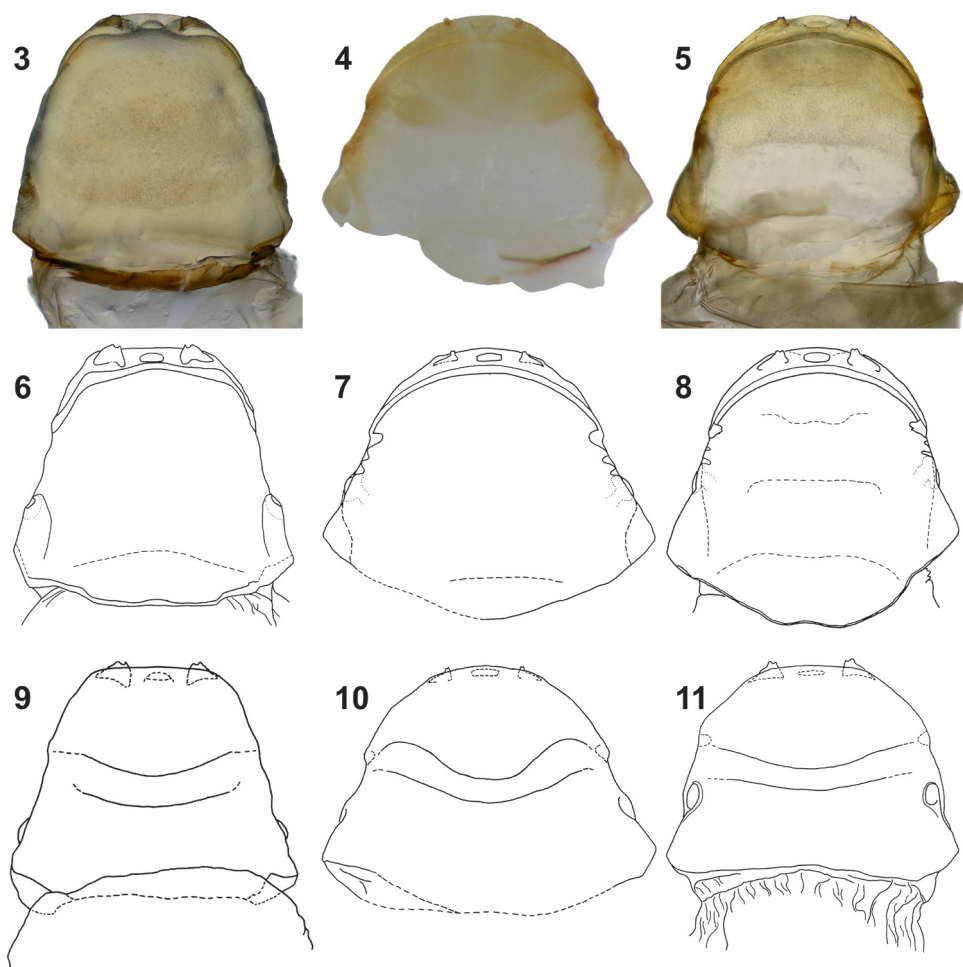
Coxal teeth are usually divided into three to five tips; coxal bristle is divided into four or five tips on foreleg and into two tips on middle and hind legs; this bifurcation is more extensive in comparison to *R. platicephala* sp. n. Coxa and trochanterofemur with more cuticular outgrowths in comparison to *R. platicephala* sp. n.

Description. Female. Canalis prolifer on abdominal segment I–IV. Tuba prolifera on segment IV positioned in posterior half of segment.

Female puparium. Cephalothorax slightly wider than long, approx. 0.7 mm long and approx. 0.8 mm wide between spiracular corners. Head wide, approx. 0.5 mm; mandible large, projecting from head contour, intermandibular distance 0.16–0.17 mm, mandibles approx. two mandibular diameters apart or less; labral apex between mandibles straight; oral ridge well developed; labral area very short; maxilla indistinct, but maxillary area with weak transverse elevation; brood opening wide, nearly from side to side, slightly sinuous, produced forward medially; head corners narrow, laterally turned posteriorly; posterolateral margin of head corner with weak apodeme; cephalic ridge well developed. Thorax without intersegmental ridges; pro-, meso- and metathorax largely fused ventrally as well as dorsally, segments seem to be subequal in length; thoracic stigma not developed; metathoracic ridge distinct, touching cintum and going up spiracle. Spiracular corners weakly prominent, obtuse; spiracula positioned anteriorly to spiracular corners, turned laterally; basal band distinct, arcuate, projecting forward, but anterior end not sharply delimited. Cephalothorax distinctly and uniformly light pigmented, only metathorax pale and translucent ventrally.

First instar. Total length (without caudal setae) 0.160–0.180 mm (n=6) on average; length of caudal setae up to 0.221 mm; ratio of body length and length of caudal seta 0.74–0.96. Ratio of body length and width approx. 2.2–2.3.

Head: Head dorsally with seven pairs of setae; ventrally strongly reduced, with setae on maxillae; mandibles and labrum overlapping outline of body; labrum not



Figures 3–11. Female puparium, cephalothorax. **3, 6, 9** *Rozenia calliopsidis* sp. n., ventral and dorsal view **4, 7, 10** *Rozenia peruana* sp. n., ventral and dorsal view **5, 8, 11** *Rozenia platicephala* sp. n., ventral and dorsal view.

emarginated; labiomaxillary area occupying majority of ventral part of head, rounded, acute posteriorly.

Thorax: Each segment of thorax bears two pairs of setae dorsally and laterally close to posterior margin, forming submedian and lateral rows of setae (Figure 20). Posterior margins of thoracic tergites smooth. Coxae broad and ovate; three coxal teeth at anterior part of each coxa, all variably divided into two to four tips; coxal bristle variably divided into four or five tips on fore leg and extensively bifid on mid and hind legs; single cuticular outgrowth positioned medially from coxal bristle; five cuticular outgrowths laterally from coxal teeth and one very short seta above cuticular outgrowths; one very short seta at the posterior part of coxa. Each trochanterofemur with spur bifid at tip,

five to six cuticular outgrowths and one short seta anteriorly and posteriorly on femur. Each tibia with five tibial spurs and short projections at distal end of tibiae. Tarsi of fore and mid legs enlarged and elongated, tarsus of hind leg rod-like and elongated. Sternal plates broad and smooth on surface and with fringe of long spinulae at its posterior margin. Precoxal pleural membrane with transverse row of microtrichia on prothorax and with two processes laterally and medially on mesothorax and metathorax.

Abdomen: Abdomen with rows of setae dorsally and laterally similar to those present on thorax (Figure 22); submedian row of setae from abdominal tergite I to tergite VIII; lateral row of setae up to tergite IX. Abdominal segment X extremely shortened and fused to segment IX; segment XI split in two parts and restricted on ventral base of caudal setae; segment XI with one particularly long caudal seta and short lateral caudal seta. Posterior margin of abdominal tergites smooth except for a few spinulae (up to six) laterally, few setae present laterally as well as mesally from lateral row of setae; posterior margin of sternites with spinulae, segment IX with only two long spinulae, which extend body outline; no spinulae immersed.

Etymology. Name derived from the generic name of the host bee.

Distribution. Argentina and Chile.

***Rozenia peruana* sp. n.**

<http://zoobank.org/84BFDD49-A710-48F1-A304-A4CA92138DCC>

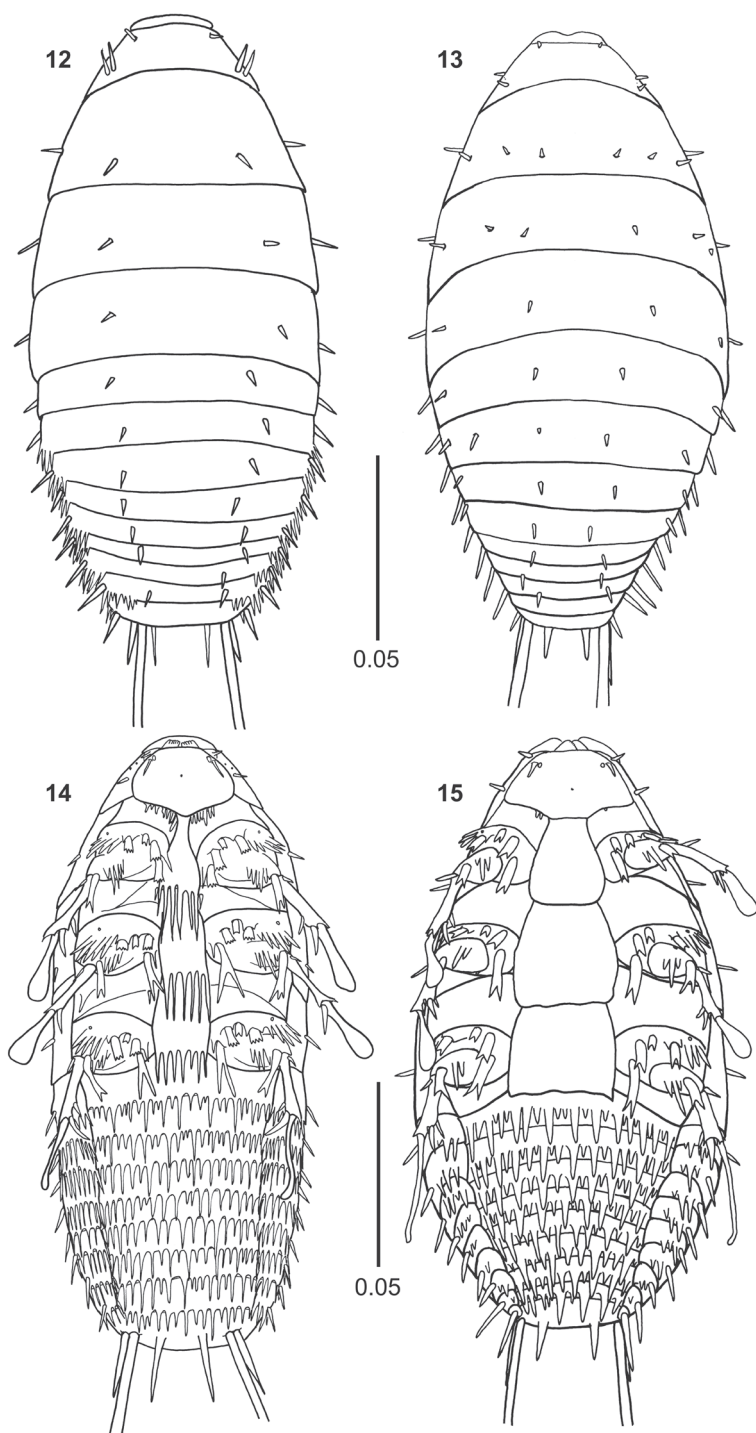
Figures 4, 7, 10

Material examined. Holotype female puparium, in a separate microvial on same pin as host. Original label: “PERU: Lima dept. / Ricardo Palma, V-9-96 / J. G. Rozen, A. Ugarte”. 1 FP, host: *Acamptopoeum vagans* (Cockerell, 1926), ♀, JS Ascher det., AMNH coll. (code: AMNH_BEE 00026923).

Differential diagnosis. Female puparium. Cephalothorax of *R. peruana* sp. n. strongly diverging posteriorly behind head (Figure 10). Among all the species of the genus *R. peruana* sp. n. has the smallest mandibles that, as in other species, project from the head contour. A very specific character is the shape of prothorax in dorsal view. Prothorax produced forward on lateral sides to the head margin, thus posterior head margin is sinuous (Figure 10). Prothorax is dorsally pigmented as in *R. platicephala* sp. n.

Description. Female. Canalis prolifer on abdominal segment I–IV, with three large tuba prolifera on segments II–IV, tuba prolifera on segment I distinct, but very small and possibly not functional.

Female puparium. Cephalothorax slightly wider than long, approx. 0.7 mm long and approx. 0.9 mm wide between spiracular corners. Head wide, approx. 0.6 mm; mandible small, projecting from head contour, intermandibular distance 0.16 mm, mandibles nearly three mandibular diameters apart; labral apex between mandibles slightly arcuate; oral ridge well-developed; epipharynx weakly divided from very short labral area; maxilla indistinct; brood opening wide, nearly from side to side, arcuate;



Figures 12–15. First instars, dorsal and ventral view. **12, 14** *Rozenia calliopsidis* sp. n. **13, 15** *Rozenia platicephala* sp. n.

head corners narrow, directed posterolaterally; posterolateral margin of head corner with distinct apodeme; cephalic ridge weak. Thorax without intersegmental ridges; pro-, meso- and metathorax largely fused ventrally, segments seem to be subequal in length; prothorax dorsally slightly shorter than half length of fused meso- and metathorax, prothorax strongly produced forward laterally; metathorax as well as mesothorax laterally with remnant of stigmata; metathoracic ridge ill-defined, but distinct, touching cintum and going anterolaterally to spiracle. Spiracular corners prominent, triangular, well-developed; spiracle positioned anterior to spiracular corners, turned laterally; basal band arcuate, projecting forward, but ill-defined. All cephalothorax pale, head, prothorax dorsally, sides of thorax and spiracular corner light pigmented; rest of cephalothorax pale and translucent.

Etymology. Name derived from the country, where the holotype was collected.

Distribution. Peru.

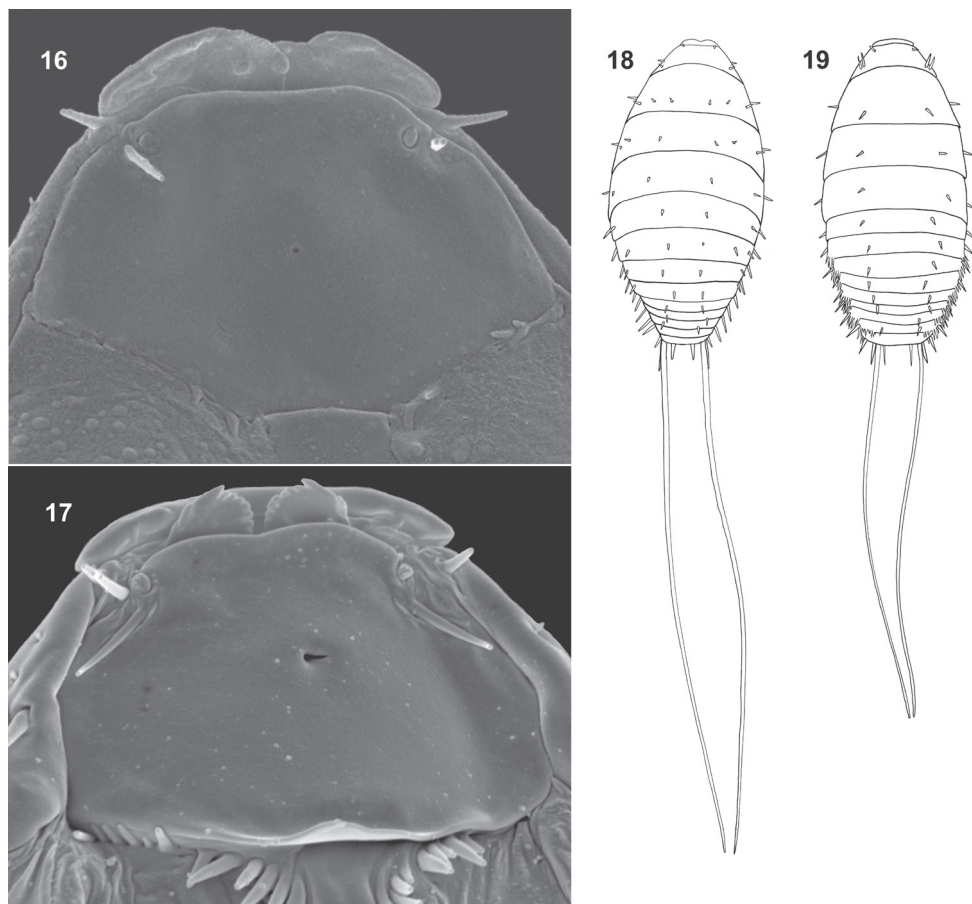
***Rozenia platicephala* sp. n.**

<http://zoobank.org/45297119-B4EB-407D-85A1-730D9A514903>

Figures 2, 5, 8, 11, 13, 15, 16, 18, 21, 23

Material examined. Holotype female puparium, in a separate microvial on same pin as host. Original label: “CHILE: Cautin Prov. / Cunco, II-1998, / Perez de Arce”. 1 FP, host: *Acamptopoeum submetallicum* (Spinola, 1851), ♀, JS Ascher det., AMNH coll. (code: AMNH_BEE 00037984).

Paratypes: ARGENTINA: Córdoba prov.: Parral, Fundo Malcho, xi.1956, 1FP, host: *A. s.* ♂, LE Peña leg., KUNHM coll. (SEMC1006814). CHILE: same as holotype, >500 L1; Limarí prov., 19 km ENE Samo Alto, 10.xi.1992, 1FP, host: *A. s.* ♀, JG Rozen, Sharkov and Snyder leg., AMNH coll. (AMNH_BEE 00037983); Cautin prov.: Cunco, ii.1998, 1 FP, host: *A. s.* ♀, Perez de Arce leg., AMNH coll. (AMNH_BEE 00037985); Bio Bío prov.: Antuco, nr. Hydroeléctrica, 37°23'49"S, 71°27'21"W, 14.xii.2004, 1 FP, host: *A. s.* ♂, JS Ascher leg., AMNH coll. (AMNH_BEE 00037986), ditto, 2 FPP, host: *A. s.* ♀, JS Ascher leg., AMNH coll. (AMNH_BEE 00037391); Dichato, 20.xii.1953, 1 FP, >500 L1, host: *A. s.* ♀, LE Peña leg., KUNHM coll. (SEMC1006914); Coquimbo prov.: Las Breas, 23.–24.x.1989, 1 FP, host: *A. s.* ♀, JG Rozen leg., AMNH coll. (AMNH_BEE 00037393); Santiago prov.: El Manzano, Valle Rio, Maipo, 1000–1500 m, i.1984, 1 FP, host: *A. s.* ♀, LE Peña leg., AMNH coll. (AMNH_BEE 00037394), El Manzano, Quebrada, 900–1500 m, 5.–6.ii.1983, 2 FPP, host: *A. s.* ♀, LE Peña leg., AMNH coll. (AMNH_BEE 00037395); Valdivia prov.: Valdivia, 9.ii.1953, 1 FP, host: *A. s.* ♀, collector not indicated, KUNHM coll. (SEMC1006957); Valparaíso prov.: Viña del Mar, La Quinta Vergara, 18.xii.2004, 1 FP, host: *A. s.* ♀, JS Ascher leg., AMNH coll. (AMNH_BEE 00037397). Other material examined: Coquimbo prov.: Las Breas, 23.–24.x.1989, 1 EMP, host: *A. s.* ♂, JG Rozen leg., AMNH coll. (AMNH_BEE 00037392); Araucanía prov.: Malleco, Victoria, xii.1985, 1 EMP, host: *A. s.* ♀, LE Peña leg., AMNH coll. (AMNH_BEE 00037396). All hosts identified by JS Ascher.



Figures 16–19. First instars, ventral view to head and dorsal view of total body. **16, 18** *Rozenia plati-cephala* sp. n. **17, 19** *Rozenia calliopsidis* sp. n.

Diagnosis. Female puparium. This species possess relatively and also absolutely the widest head among all species of the genus. Spiracular corners are sharply triangular and distinctly prominent, but not large. Prothorax is more pigmented dorsally than other parts of thorax, like in *R. peruana* sp. n., but anterior and posterior margins are paralel, arcuate, producing forward on sides only slightly. Position of spiracula seems to be characteristic for this species. They are turned more dorsally than in other species, however this character is very variable and may be inconsistent. Pigmentation like in *R. peruana* sp. n.

First instar. Shape of body is more rounded than in *R. calliopsidis* sp. n., width of segments decreases from metathorax more strongly. Ratio of body length and width is on average 2.0. Ratio of body length and length of caudal setae is approx. 0.60–0.65. Caudal setae are relatively longest among all species.

Head dorsally with six pairs of setae compared to seven and usually longer in *R. calliopsidis* sp. n.; labrum is narrow at the middle contrary to *R. calliopsidis* sp. n.; labium is projecting more laterally than in *R. calliopsidis* sp. n.

Each segment of thorax bears four pairs of setae dorsally and laterally, forming submedian, interstitial, supralateral and lateral rows of setae. Sternal plates are broad and smooth on surface, specific are also smooth posterior margins. Precoxal pleural membrane is smooth without any projections except of one or two cuticular outgrowths on prothoracic precoxal pleural membrane. Coxal teeth are always bifid in two tips in contrast to *R. calliopsidis* sp. n. with as many as five tips; coxal bristle is always divided into two tips and bifurcation in middle leg and hind leg is not so extensive, there are no cuticular outgrowth by coxal bristle contrary to *R. calliopsidis* sp. n., also there are not so many cuticular outgrowths on coxa and femur like in *R. calliopsidis* sp. n.

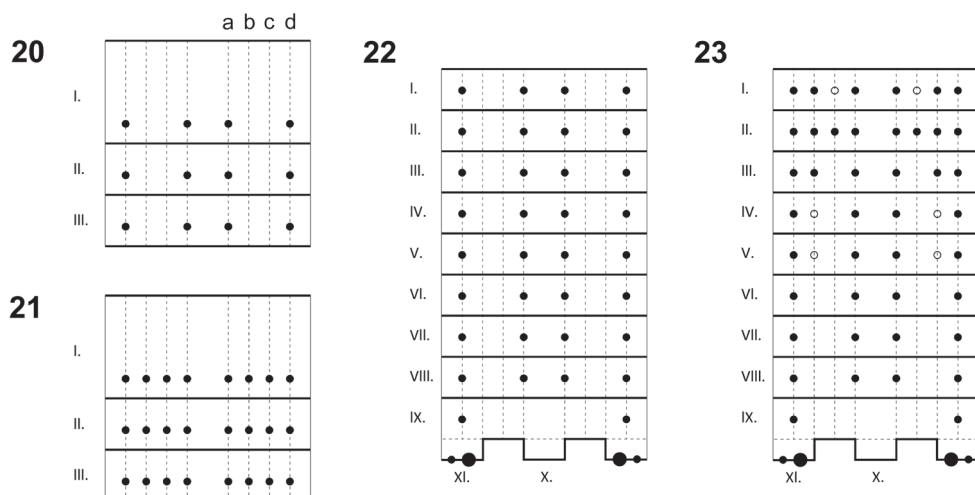
All four pairs of rows of setae continues dorsally on abdomen, submedian row up to tergite XIII, interstitial row is on tergite II or in some specimens also on tergite I, supralateral row is variable and reach up to tergite III, IV or V, and lateral row up to tergite IX. Spinulae on posterior margins of abdominal tergites only beyond lateral row and not visible in dorsal view.

Description. Female. Canalis prolifer on abdominal segment I–IV. Tuba prolifera on segment IV positioned in posterior half of segment.

Female puparium. Cephalothorax slightly wider than long, approx. 0.8 mm long and approx. 0.9–1.0 mm wide between spiracular corners. Head wide, approx. 0.7 mm; mandible projecting from head contour, intermandibular distance 0.17–0.21 mm, approx. two mandibular diameters apart, but variable among different individuals; labral apex between mandibles straight; oral ridge well developed; epipharynx weakly divided from labral area, short; maxilla not developed, but maxillary area with weak transverse elevation; brood opening wide, nearly from side to side, arcuate; head corners narrow, directed posterolaterally; posterolateral margin of head corner with distinct apodeme; cephalic ridge weak. Thorax without intersegmental ridges; pro-, meso- and metathorax largely fused ventrally, segments seem to be subequal in length, prothorax dorsally slightly shorter than half length of fused meso- and metathorax; meso- and metathorax laterally with remnants of stigma, mesothoracic spiraculum very small and hardly visible; metathoracic ridge ill-defined, but distinct, touching cinctum and going up spiracle. Spiracular corners prominent, well developed; spiracula positioned anterior to spiracular corners, turned dorsally; basal band distinct but weak, arcuate, projecting forward. All cephalothorax pale, head and prothorax dorsally and head, prothorax and mesothorax light pigmented ventrally; spiracular area and basal band only slightly darker; rest of cephalothorax pale and translucent.

First instar. Total length approx. 0.154–0.175 mm (n=3) without caudal setae; length of caudal setae up to 0.289 mm (on an average 0.276 mm); ratio of body length and length of caudal setae approx. 0.60–0.65. Ratio of body length and width approx. 1.9–2.3.

Head: Head dorsally with six pairs of setae; ventrally strongly reduced; with setae on maxillae; distinctive mandibles and labrum overlapping outline of body; labrum emarginated; labiomaxillary area occupying majority of ventral part of head, rounded, posterior margin straight.



Figures 20–23. Diagram of first instar chaetotaxy. **20, 22** *Rozenia calliopsidis* sp. n., thoracic and abdominal tergites; **21, 23** *Rozenia platicephala* sp. n., thoracic and abdominal tergites; a-submedian row of setae, b-interstitial row of setae, c-supralateral row of setae, d-lateral row of setae; point-stable presence of seta; circle-seta with unstable presence; large point-caudal seta.

Thorax: Each segment of thorax bears four pairs of setae dorsally and laterally close to posterior margin forming submedian, interstitial, supralateral, and lateral rows of setae (Figure 21). Posterior margins of thoracic tergites smooth. Coxae broad and ovate; on each coxa three coxal teeth and one coxal bristle at anterior part of coxa, all bifid at tips; three or four cuticular outgrowths laterally from coxal teeth and one very short seta above cuticular outgrowths and one on posterior margin of coxa. Each trochanterofemur with femoral spur bifid at tip; two or three cuticular outgrowths and one short seta anteriorly and posteriorly on femur. Each tibia with five tibial spurs and little projections at distal end of tibiae. Tarsi of fore and middle legs enlarged and elongated, tarsi of hind legs rod-like and elongated. Sternal plates broad and smooth on surface and on posterior margins. Precoxal pleural membrane smooth without any projections except of one or two cuticular outgrowths on prothoracic precoxal pleural membrane.

Abdomen: Abdomen with rows of setae dorsally and laterally similar to those present on thorax; submedian row from abdominal tergite I to tergite VIII; interstitial row on tergite II or in some specimens also on tergite I; supralateral row variable up to tergite III, IV or V; lateral row up to tergite IX (Figure 23). Abdominal segment X extremely shortened and fused to segment IX; segment XI split in two parts and restricted only on ventral base of caudal setae; segment XI with particularly long caudal seta and short lateral caudal seta. Posterior margin of abdominal tergites smooth except for lateral part with a few spinulae (up to three) more laterally than lateral row of setae; posterior margin of sternites with spinulae, segment IX with only two longer spinulae, which extend body outline; no spinulae immersed.

Etymology. Name of this species refers to characteristic flat head and general flat appearance of all *Rozenia* gen. n. species, when found between tergites of host bees.

Distribution. Argentina and Chile.

Published hosts assigned to *R. platicephala* sp. n. *A. submetallicum*: Schwarz (1931: 78-79), record from Chile (as *Liopoeum submetallicum* (Spinola)), also reported by Hofeneder and Fulmek (1943: 35), but with no original data.

Note: To *R. platicephala* sp. n. could be assigned findings of Strepsiptera in the host bee *Acamptopoeum argentinum* (Friese, 1906): Hofeneder and Fulmek (1943: 42), record from Argentina (as *Perdita argentina* Friese), repeated by Hofeneder (1949: 122) and later by Hofeneder (1952: 489) (as *Calliopsis (Parafriesea) argentina* (Friese)). The record is impossible to verify as reliable pending a review of the material. The information about material deposition is not known to us.

Key to species of the genus *Rozenia* gen. n.

Female puparia and females

- 1a More than four abdominal segments developed, with tuba prolifera III (if developed) positioned in the middle part of abdominal segment IV; combination of characters different **other Strepsiptera**
- 1b Only four abdominal segments developed, with tuba prolifera III positioned on the posterior half of abdominal segment IV (Figures 1–2); brood opening wide, almost from side to side, about four times wider than intermandibular distance, or more; head wider than distal part of prothorax, this character cause that side of cephalothorax is not continuously diverging posteriorly; mandibles extending from the head contour in ventral view; intersegmental ridges not developed (Figures 3–11) **2, *Rozenia* gen. n.**
- 2a Spiracular corners weakly prominent, obtuse, not triangular; brood opening turned backwards laterally, very close to posterior margin of mandible and continued as cephalic ridge (Figures 3, 6); cephalothorax pigmented in all parts (Figure 3); host bee *Calliopsis* spp. ***Rozenia calliopsidis* sp. n.**
- 2b Spiracular corners prominent, triangular; brood opening fluently transforms into cephalic ridge and forms an arcuate line (Figures 4, 5, 7, 8); posterior half of cephalothorax nearly transparent (Figures 4, 5); host bee *Acamptopoeum* spp. **3**
- 3a Prothorax dorsally produced forward on lateral sides to the head margin, thus posterior head margin is sinuous (Figure 10); mandibles very small (Figures 4, 7) ***Rozenia peruana* sp. n.**
- 3b Anterior and posterior margins of prothorax dorsally parallel, thus posterior head margin arcuate (Figure 11); mandibles of normal size (Figures 5, 8) ***Rozenia platicephala* sp. n.**

First instars

- 1a Submedian row of setae absent on abdomen; caudal setae shorter or as long as body; posterior margin of labium emarginated; ventral sublateral bristle on sternite IX; posterior margin of thoracic tergites with spinulae..... **other Strepsiptera**
- 1b Submedian row of setae present on abdomen; caudal setae longer than body; posterior margin of labium continuous; ventral sublateral bristle absent; posterior margin of thoracic tergites smooth **2, *Rozenia* gen. n.**
- 2a Sternal plates at posterior margin with spinulae; interstitial and supralateral row of setae on dorsum absent; coxal tooth with two to four tips at apex; coxal bristle in fore leg with multiple tips at apex; coxal bristles in mid and hind leg extensively bifid; numerous cuticular outgrowths on precoxal pleural membrane and coxae; caudal setae slightly longer than body ***Rozenia calliopsidis* sp. n.**
- 2b Sternal plates smooth on posterior margin; interstitial and supralateral row of setae on dorsum; coxal tooth bifid at apex; coxal bristles bifid on each leg; few cuticular outgrowths on precoxal pleural membrane and coxae; caudal setae distinctly longer than body ***Rozenia platicephala* sp. n.**

Discussion

Among all, the newly described genus, *Rozenia* gen. n., is morphologically unusual in having extremely long caudal setae in first instars. No other Strepsiptera species possess such long caudal setae (Pohl 2000). These setae are always longer than the body in *Rozenia* gen. n., and almost two times longer than the body in *R. platicephala* sp. n. (Figures 18–19). This species is also exceptional in having four rows of dorsal thoracic setae, one row more than in the most basal Strepsiptera family Mengenillidae (Pohl 2000). Until now, the chaetotaxy of first instars seemed to be reductive in the evolution of Strepsiptera, because basal lineages possess more abundant setae on dorsal part of the thorax and abdomen than derived lineages. It is, however, clear that *Rozenia* gen. n. is not related to the Mengenillidae, but belongs to the family Stylopidae, which means that at least one row of setae are newly developed in *Rozenia* gen. n. We call the new row of setae the “interstitial row”, because at most three rows of setae were known in all other Strepsiptera until now. This interstitial row continues to abdominal segments I and II in *R. platicephala* sp. n. The second species of *Rozenia* gen. n. with known first instars, *R. calliopsidis* sp. n., has a more standard chaetotaxy, but a submedian row of setae is present on the thorax, as well as on abdominal segments I–VIII, which is a synapomorphy of the genus *Rozenia* gen. n.

Rozenia gen. n. is a genus distinctive from other Strepsiptera genera in numerous characters mentioned in generic diagnosis. According to the host family and a few

shared characters, it seems to be most closely related to the genus *Crawfordia*. In both genera, a single median tuba prolifera on canalis prolifer is present on segments II–IV of the abdominal part of female puparia. In first instars, spinulae are not immersed in any part of the body; two pairs of setae or more are present on each thoracic and abdominal segment dorsally; the sternal plates are completely smooth, no setae are developed; coxal teeth, coxal bristles and femoral spurs are bifid or with multiple tips in both genera. Some of these characters are developed in some other Strepsiptera species, but never in the family Stylopidae (Pohl 2000).

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The oribatid mite genus *Benoibates* (Acari, Oribatida, Oripodidae)

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Abstract

Two species of oribatid mites of the genus *Benoibates* (Oribatida, Oripodidae), i.e., *B. bolivianus* Balogh & Mahunka, 1969(a) and *B. minimus* Mahunka, 1985, are recorded for the first time in Costa Rica. Both are redescribed in details, using drawings, images and SEM micrographs, on the basis of Costa Rican specimens. An identification key to the known species of *Benoibates* is given.

Keywords

Oribatid mites, *Benoibates*, redescription, key, Costa Rica

Introduction

Benoibates (Acari, Oribatida, Oripodidae) is a genus of oribatid mites that was proposed by Balogh (1958) with *Benoibates flagellifer* Balogh, 1958 as type species. Currently, it includes 12 species, distributed in the Neotropical region (seven species), U.S.A. (two species), Ethiopian region (two species) and Polynesia (one species) (data summarized by Subías 2004, updated 2014).

Subías (2004, updated 2014) includes additionally three genera as junior synonyms in *Benoibates*: *Exoripoda* Woolley 1961 (with two species: *E. excavatus* Woolley, 1961; *E. suramericanus* Mahunka, 1983), *Haploripoda* Balogh & Mahunka, 1967 (with one species: *H. reductus* Balogh & Mahunka, 1967) and *Reductoripoda* Mahunka & Palacios-Vargas, 1996 (with one species: *R. absoluta* Mahunka & Palacios-Vargas, 1996). *Exoripoda* was described by Woolley (1961) on bases of the presence of one pair of adanal setae (versus two pairs in *Benoibates*); *Haploripoda* – Balogh and Mahunka (1967) on bases of the presence of one pair of genital setae (versus two pairs in *Benoibates*); *Reductoripoda* – Mahunka and Palacios-Vargas (1996) on bases of the presence of one pair of adanal setae and the absence of anal setae (versus two and one pairs present in *Benoibates*, accordingly). Subías probably is right, but we cannot support his opinion at this moment, because many oripodid genera were proposed on the basis of distinctions in the number of anogenital setae – see also different views on classification of genera (for example, Woolley 1961, 1966; Aoki and Ohkubo 1974; Balogh and Balogh 1992, 1999; Subías 2004). Hence, the full and detail revision of all taxa in the family Oripodidae is necessary in the future.

The main generic characters of *Benoibates* in Oripodidae are (summarized by Balogh 1958, Aoki and Ohkubo 1974; Balogh and Balogh 1990, including our additions): rostrum rounded; bothridial openings not covered by notogaster; body surface foveolate; anterior notogastral margin convex medially, transverse straight; 10 (exception 11) pairs on notogastral setae present; two pairs of genital, one pair of aggenital, two pairs of adanal and one pair of anal setae present, genital setae inserted in anterior part of genital plates, anal setae inserted in posterior part of anal plates; legs tridactylous.

In the course of proceeding taxonomic identification of oribatid mites from Costa Rica (Ermilov et al. 2014a, b), we have found two species of *Benoibates*, *B. bolivianus* Balogh & Mahunka, 1969(a) (described from Bolivia) and *B. minimus* Mahunka, 1985 (described from Antilles). Both species are recorded for the first time in Costa Rican fauna.

The original descriptions of *B. bolivianus* and *B. minimus* were based only on holotypes, and, hence, it is incomplete and brief (lacking information about the measures of morphological structures, leg setation and solenidia, morphology of gnathosoma; only dorsal and ventral sides of body are illustrated). We also notice that *Benoibates*-species are very similar morphologically, and species descriptions of this genus were brief. Therefore their supplementary descriptions are especially important now. The main goal of our paper is to present detailed redescriptions and illustrations of *B. bolivianus* and *B. minimus*, using drawings, images and SEM (Scanning Electron Microscopy) micrographs, of Costa Rican specimens.

The second goal of our paper is to present an identification key to the based on *Benoibates* known species.

Materials and methods

Material

Benoibates bolivianus Balogh & Mahunka, 1969

Three specimens (male and two females), Costa Rica, 9°50'24"N, 83°53'17"W, Cartago, Dulce Nombre, Paraíso, Jardín Botánico Lankester, 1400 m a.s.l., in leaf litter in secondary forest, 14.V.2013, collected by O. Alvarado-Rodríguez and A.P. Retana-Salazar. Holotype (0-555-68, Hungarian National History Museum, Budapest) (see Balogh and Mahunka 1969a): Bolivia, "Guayaramerin, Beni, forest along the river Mamore, litter and wooden debris from the shady base of a low tree", 26.XI.1966 (collected by J. Balogh, S. Mahunka and A. Zicsi).

Benoibates minimus Mahunka, 1985

Five specimens (four males and one female), Costa Rica, 9°50'24"N, 83°53'17"W, Cartago, Dulce Nombre, Paraíso, Jardín Botánico Lankester, 1400 m a.s.l., in leaf litter in secondary forest, 14.V.2013, collected by O. Alvarado-Rodríguez and A.P. Retana-Salazar. Holotype (971-HO-84, Hungarian National History Museum, Budapest) (see Mahunka 1985): Antilles, "Anse La Raye, Piloni Pt., singling from under bark of coastal trees and sifting rotten material accumulated at tree bases and picking out animals", 14.VII.1980 (collected by S. Mahunka).

Methods

The specimens were mounted in lactic acid on temporary cavity slides for measurement and illustration. The body length was measured in lateral view, from the tip of the rostrum to the posterior edge of the ventral plate. The notogastral width refers to the maximum width in dorsal aspect. Lengths of body setae were measured in lateral aspect. All body measurements are presented in micrometers. Formulae for leg setation are given in parentheses according to the sequence trochanter–femur–genu–tibia–tarsus (femulus included). Formulae for leg solenidia are given in square brackets according to the sequence genu–tibia–tarsus.

General terminology used in this paper follows that of Grandjean (summarized by Norton and Behan-Pelletier 2009).

Drawings were obtained by a drawing tube using the Carl Zeiss transmission light microscope "Axioskop-2 Plus". Images were obtained by an AxioCam ICc3 camera using the Carl Zeiss transmission light microscope "Axio Lab.A1". SEM micrographs were obtained by the Jeol scanning electron microscope "JSM-6510 LV".

Results

Redescription of the studied species

Benoibates bolivianus Balogh & Mahunka, 1969(a)

Figs 1–42

Diagnosis. Body size: 514–597 × 265–332. Body surface weakly foveolate. Rostral, lamellar and interlamellar setae setiform, barbed; latter are longest. Bothridial setae short, clavate. Ten pairs of notogastral setae of medium size (24–32). Sacculi *Sa* large than other. Subcapitular setae *h* longer than *a* and *m*. Apodemes 2 connected medially and to anterior margin of genital aperture. Pedotecta II with one pointed tip anteriorly. Genital and aggenital setae short. Anal and adanal setae very long, flagellate.

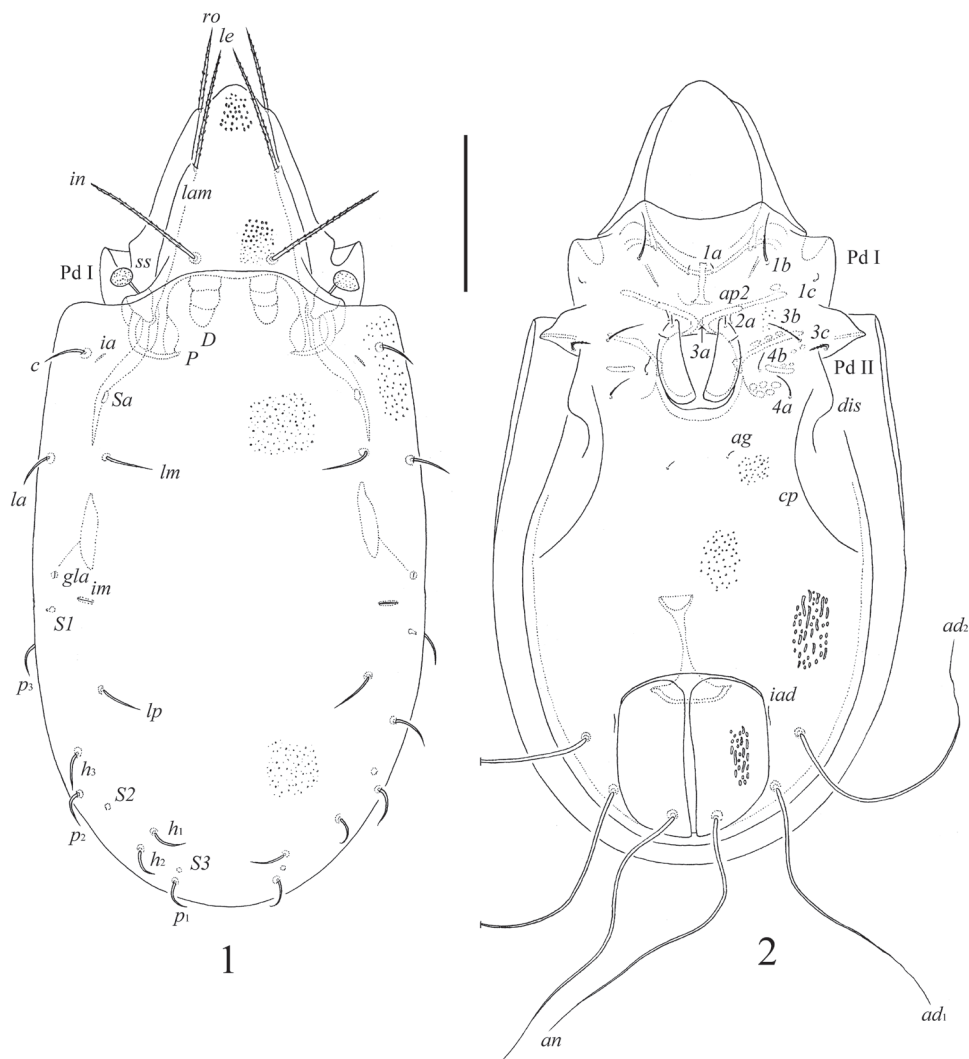
Description. *Measurements.* Body length: 514–597 (three specimens); notogaster width: 265–332 (three specimens).

Integument. Body color yellowish brown to brown. Body surface weakly foveolate: prodorsum with distinct, round foveoles, larger in antero-medial part (up to 4) than in basal part (up to 1); notogaster, epimeral region, subcapitular mentum and gena, and genital plates with weak, round foveoles (up to 2); anogenital region and legs with distinct (except weak between genital and anal apertures), round or oval foveoles (up to 4), simultaneously also with longitudinal foveoles (length up to 16). Body surface of notogaster and ventral side covered by microgranular cerotegument (less than 1; visible only high magnification, ×1000).

Prodorsum. Rostrum weakly protruding, rounded. Lamellae (*lam*) located dorso-laterally, half length of prodorsum (measured in lateral view), without cusps. Translamella absent. Prolamellar lines (*plam*) thin, reaching the insertions of rostral setae and bend ventrally to meet the rostral margins. Sublamellar lines (*slam*) distinct, long. Sublamellar porose areas (*Al*) small, rounded (4–6). Keel-shaped ridges (*kf*) well developed. Rostral (*ro*, 55–61), lamellar (*le*, 77–86) and interlamellar (*in*, 94–106) setae setiform, barbed. Interlamellar setae long, reaching the insertions of lamellar setae. Bothridial setae (*ss*, 32–41) with short stalk (16–21) and clavate, barbed head (16–20). Exobothridial setae (*ex*, 16) thin, smooth.

Notogaster. Anterior notogastral margin weakly convex, trapezoid. Dorsophragmata (*D*) elongated, not reaching pleurophragmata (*P*). Notogastral shoulders rectangular in dorsal view, anterior margin almost transverse straight. Ten pairs of notogastral setae of medium size (24–32), setiform, indistinctly barbed (visible under high magnification, ×1000). Insertions of setae h_1 – h_3 varies. Four pairs of sacculi developed: *Sa* largest, located postero-medially to setae *c*; *S1* – postero-laterally to lyrifissures *im*; *S2* – between setae h_2 and h_3 ; *S3* – anteriorly to p_1 . Lyrifissures *ia* located medially to setae *c*; *im* – between setae *lm* and *lp*, in transverse position; *ip* – laterally to p_1 ; *ih* – anteriorly to p_1 ; *ips* – between p_2 and p_3 . Opisthonotal gland openings (*gla*) located antero-laterally to lyrifissures *im*.

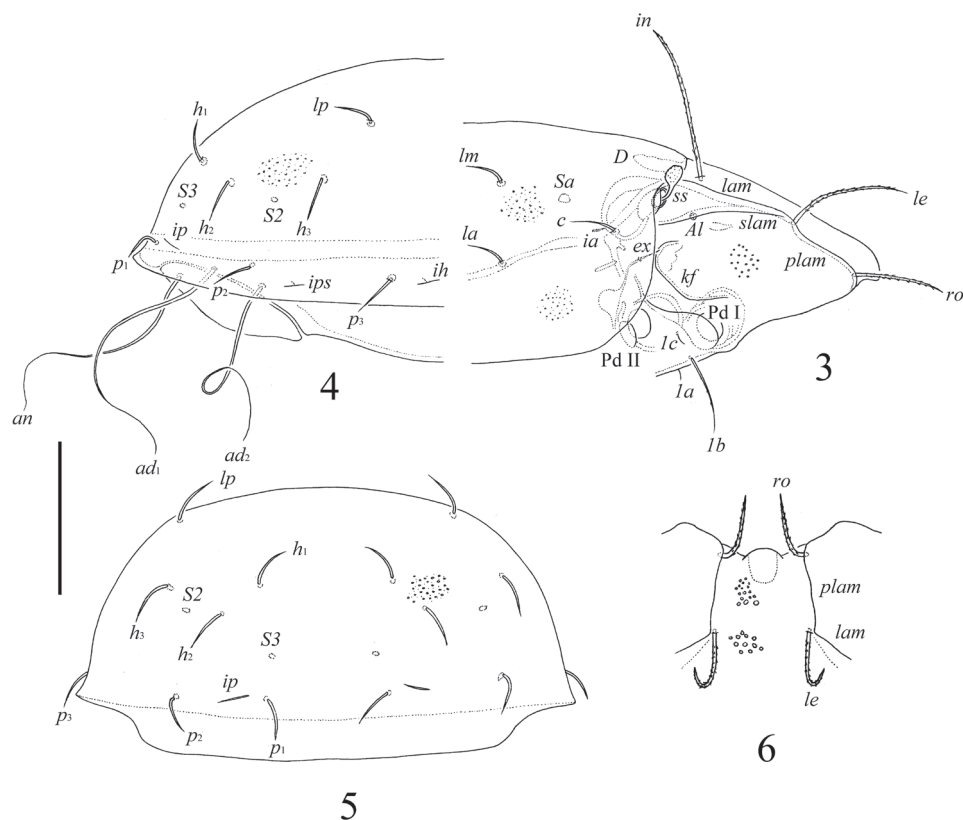
Gnathosoma. Subcapitulum longer than wide (118–127 × 86–98). Subcapitular setae setiform, slightly barbed; *h* (53–61) longer than *a* and *m* (both 28–32). Setae *m*



Figures 1–2. *Benoibates bolivianus* Balogh & Mahunka, 1969, Costa Rican specimen: **1** dorsal view **2** ventral view (gnathosoma and legs not illustrated). Scale bar 100 μ m.

thinnest. Two pairs of adoral setae (or_1 , or_2 , 20) setiform, densely ciliate. Palps (length 77) with setation 0–2–1–3–9(+ ω). Solenidion attached to eupathidium. Chelicerae (length 127–139) with one barbed setae (cha , 36–41), chb and their alveoli absent. Trägårdh's organ (Tg) long, tapered.

Epimeral and lateral podosomal regions. Apodemes 1, 2, 3, sejugal and sternal apodemes distinct. Apodemes 2 ($ap2$) connected medially and to anterior margin of genital aperture. Sternal apodeme of medium size, not reaching the apodemes 2. Epimeral setal formula: 3–1–3–2. Centrovventral setae $1a$, $2a$, $3a$ smooth, other slightly barbed; $1b$ (41–53) longer than $3b$ (24–28), $4a$ (20), $4b$, $3c$ (16–18), $1c$, $2a$ (10) and $1a$, $3a$

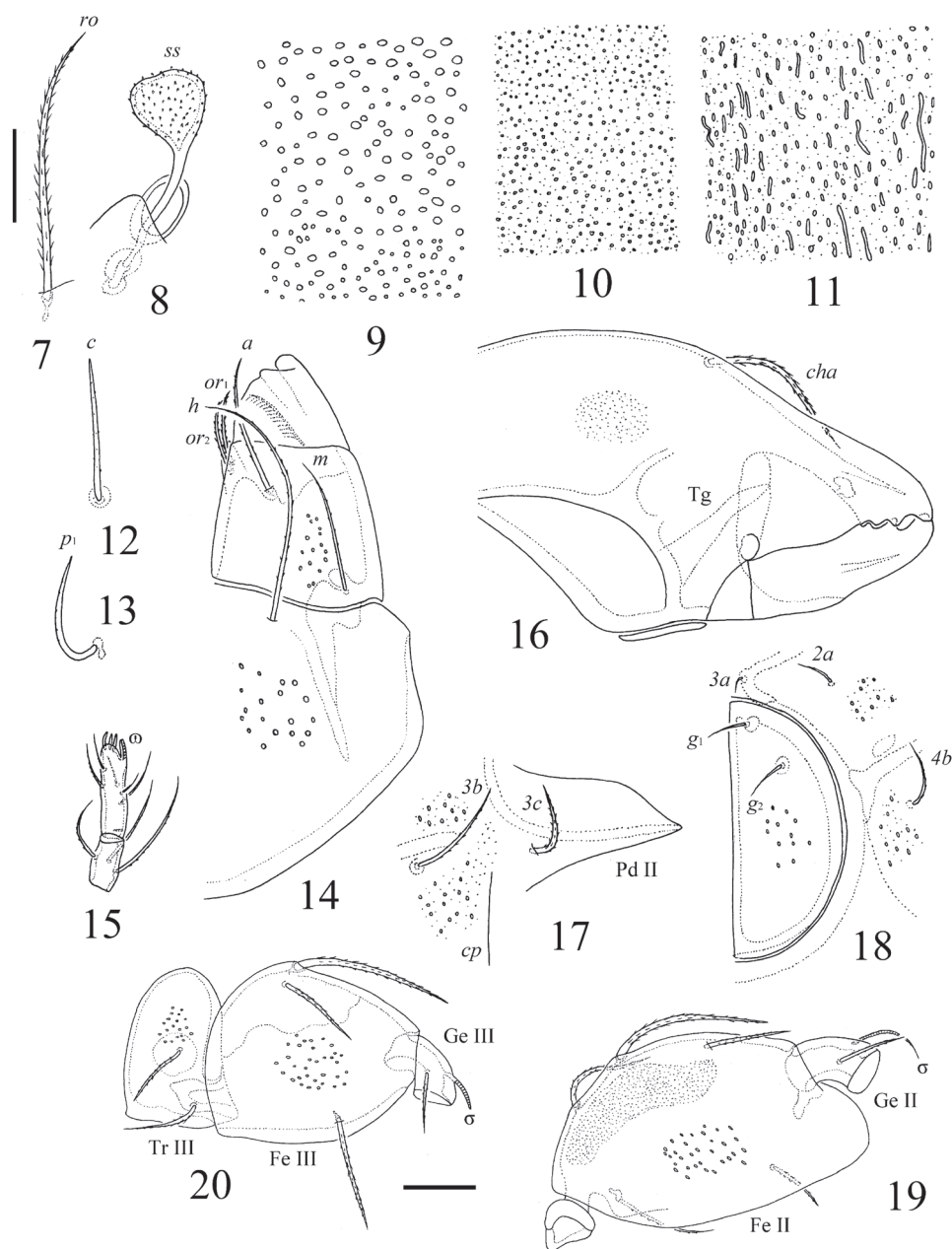


Figures 3–6. *Benoibates bolivianus* Balogh & Mahunka, 1969, Costa Rican specimen: **3** lateral view of prodorsum and anterior part of notogaster and pteromorph (gnathosoma and legs I, II not illustrated) **4** lateral view of notogaster **5** posterior view of notogaster **6** frontal view of prodorsum. Scale bar 100 μ m.

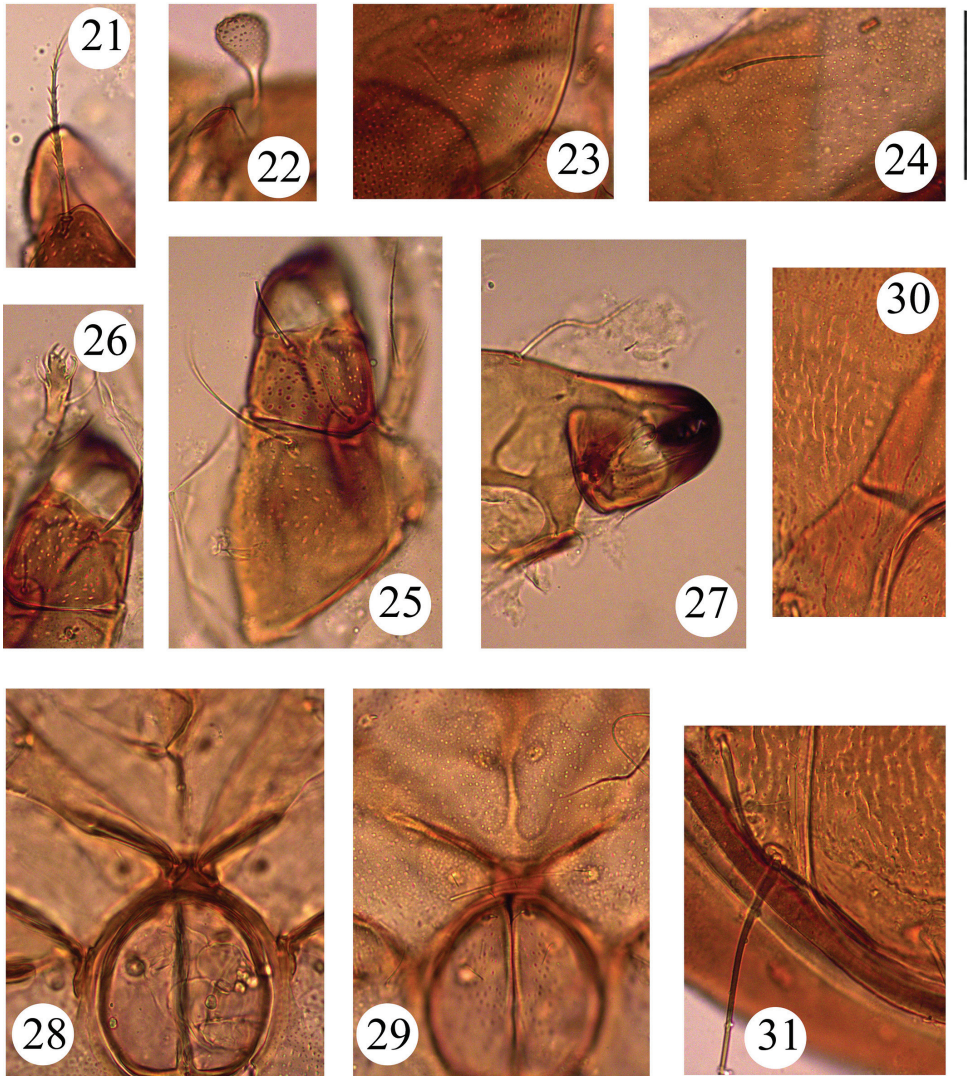
(6–8). Setae $3c$ thickest. Pedotecta I (Pd I) large, concave (measured in dorsal view) and scale-like (measured in lateral view); pedotecta II (Pd II) smaller, trapezoid, with one pointed tip anteriorly (measured in ventral view) and scale-like (measured in lateral view). Discidia (*dis*) elongated, weakly triangular. Circumpedal carinae (*cp*) distinct.

Anogenital region. Two pairs of genital (g_1 , g_2 , 10) and one pair of aggenital (*ag*, 8) setae setiform, thin, smooth. One pair of anal (*an*) and two pairs of adanal (ad_1 , ad_2) setae (all 176–196) very long, flagellate. Lyrifissures *iad* located close to and parallel anal plates.

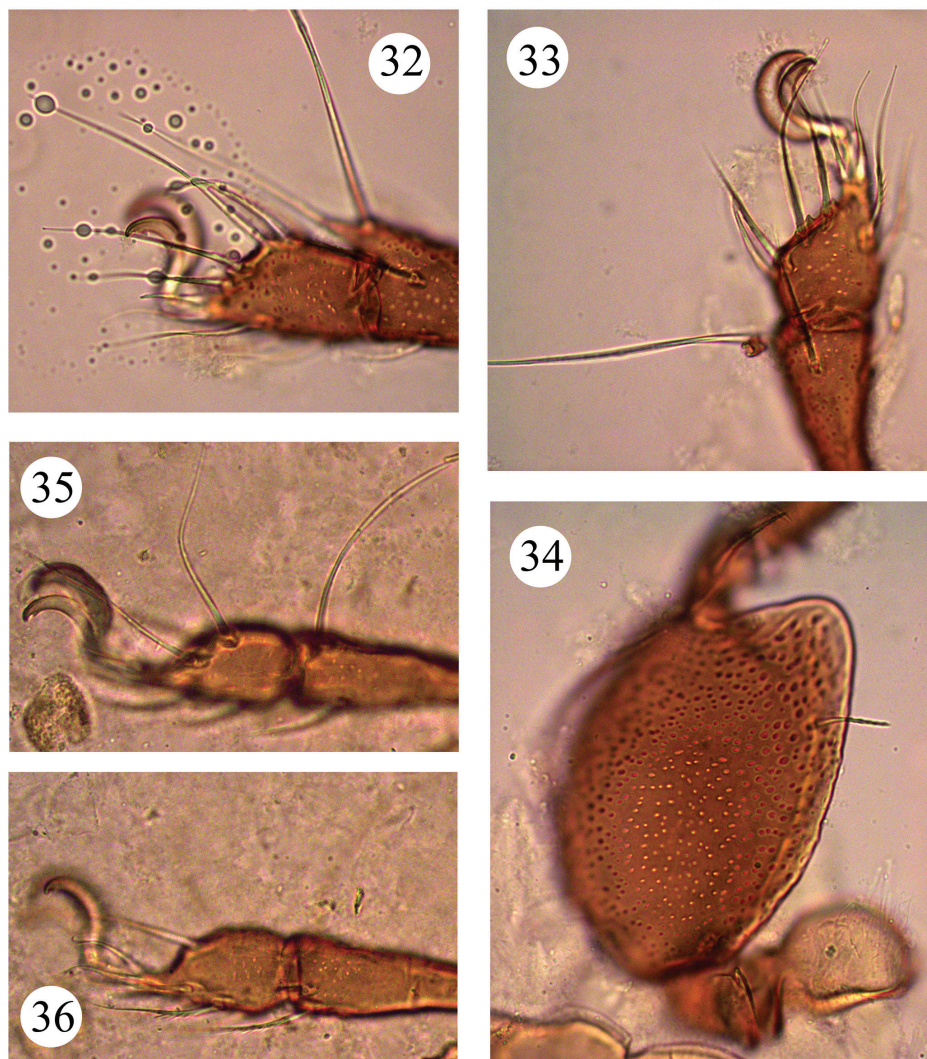
Legs. Median claw weakly thicker than two lateral claws; all with several minute barbs on dorsal side. Lateral claws with ventral tooth. Formulae of leg setation and solenidia: I (1–5–2–4–16) [1–2–2], II (1–5–2–4–13) [1–1–2], III (2–3–1–3–13) [1–1–0], IV (1–2–2–3–11) [0–1–0]; homology of setae and solenidia indicated in Table 1. Famulus short, straight, slightly dilated distally, truncated. Solenidia ω_2 on tarsi I, ω_1 and ω_2 on tarsi II, σ on genua II, III of medium size, thickened, blunt-ended. Other solenidia long, setiform.



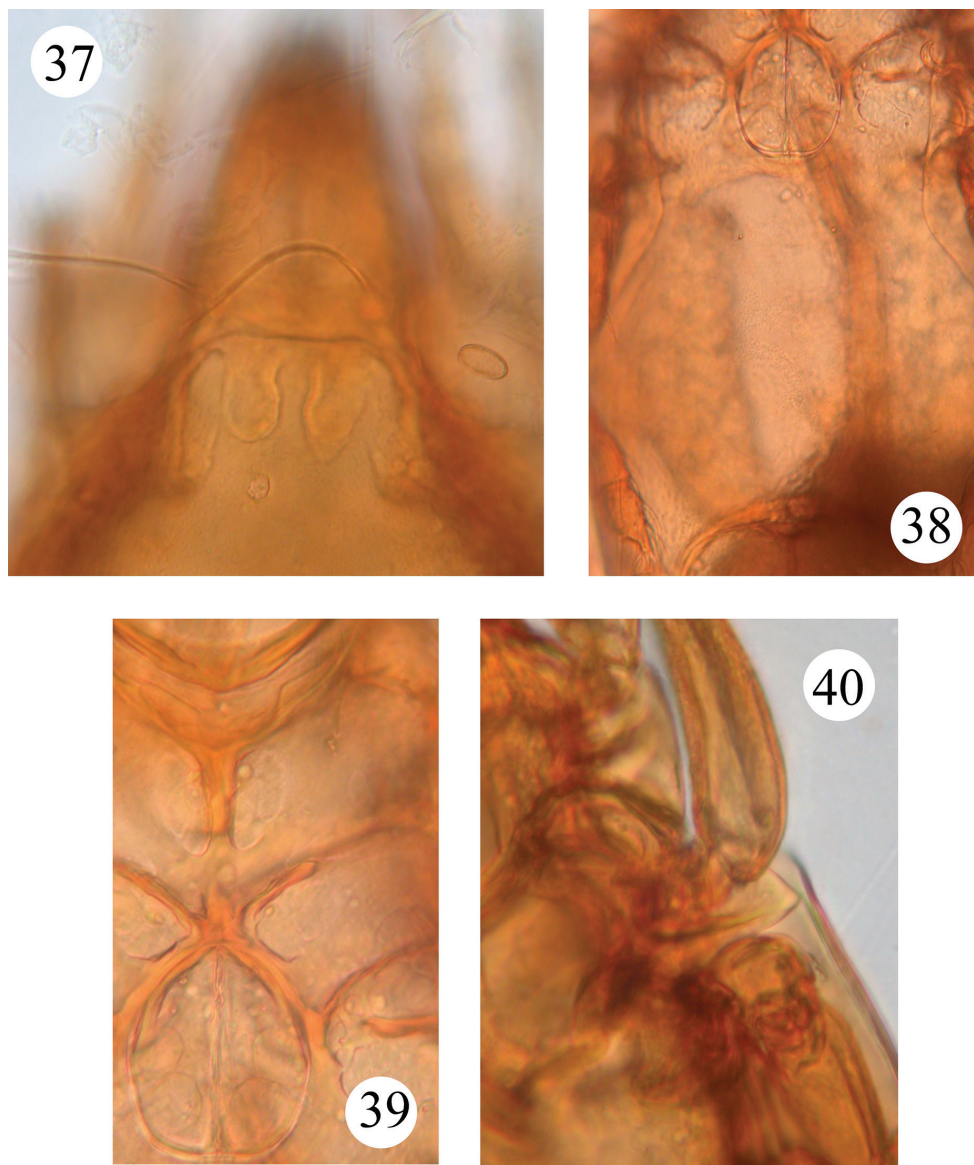
Figures 7–20. *Benoibates bolivianus* Balogh & Mahunka, 1969, dissected Costa Rican specimen: **7** rostral seta **8** posterior bothridial seta **9** foveoles in medio-basal part of prodorsum **10** foveoles in central part of notogaster **11** foveoles in lateral part of anogenital region **12** notogastral seta **13** notogastral seta p_1 **14** left part of subcapitulum, ventral view **15** tarsus and tibia of palp **16** antero-medial part of chelicera **17** pedotectum II, anterior part of circumpedal carina and epimeral setae **3b**, **3c** **18** left genital plate and epimeral setae **2a**, **3a**, **4b** **19** femur and genu of left leg II, paraxial view **20** trochanter, femur and genu of left leg III, antiaxial view. Scale bar 20 μ m.



Figures 21–31. *Benoitibates bolivianus* Balogh & Mahunka, 1969, dissected Costa Rican specimen, microscope images: **21** rostral seta **22** bothridial seta **23** foveoles in anterior part of pteromorph **24** notogastral seta h_3 **25** left part of subcapitulum, ventral view, and medio-basal part of left palp **26** right rutellum and gena of subcapitulum, ventral view, and anterior part of right palp **27** antero-medial part of chelicera **28** genital plates and central part of epimeral region **29** genital plates and central part of epimeral region **30** antero-lateral part of right anal plate, insertion of adanal setae ad_2 , and foveoles in anogenital region **31** posterior part of right anal plate and insertions of anal and adanal setae. Scale bar 50 μm .



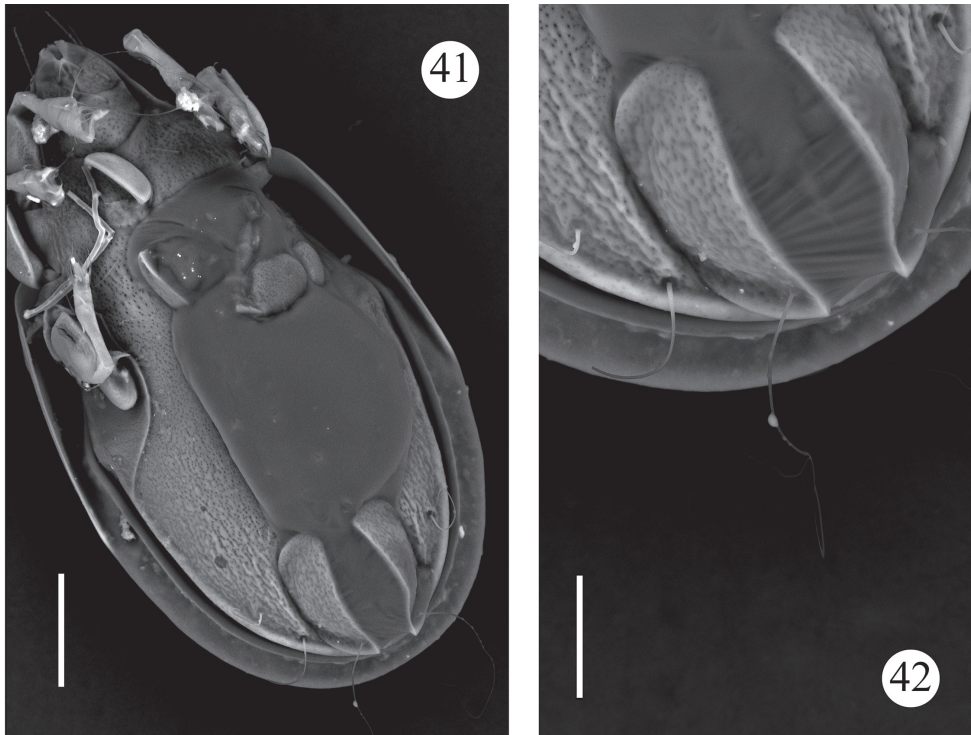
Figures 32–36. *Benoibates bolivianus* Balogh & Mahunka, 1969, dissected Costa Rican specimen, microscope images: **32** tarsus and anterior part of tibia of leg I, left, anti-axial view **33** tarsus and antero-medial part of tibia of leg II, right, anti-axial view **34** basal part of tibia, genu, femur and trochanter of leg II, right, anti-axial view **35** tarsus and antero-medial part of tibia of leg III, right, anti-axial view **36** tarsus and antero-medial part of tibia of leg IV, right, anti-axial view. Scale bar 50 µm.



Figures 37–40. *Benoitibates bolivianus* Balogh & Mahunka, 1969, holotype, microscope images: **37** dorsal view of prodorsum and anterior part of notogaster **38** ventral view of anogenital region **39** genital plates and central and left parts of epimeral region **40** ventral view of left pedotectum II.

Remarks. Costa Rican specimens of *Benoitibates bolivianus* are similar in all morphological characters to Bolivian specimens from the original description (Balogh and Mahunka 1969a), except slightly shorter epimeral setae *1b*.

Distribution. Neotropical region.



Figures 41–42. *Benoibates bolivianus* Balogh & Mahunka, 1969, Costa Rican specimen, SEM micrographs: **41** ventral view **42** ventral view of ano-adanal region. Scale bar 100 μm (**41**), 50 μm (**42**).

Table 1. Leg setation and solenidia of *Benoibates bolivianus* Balogh & Mahunka, 1969 (same data for *B. minimus* Mahunka, 1985).

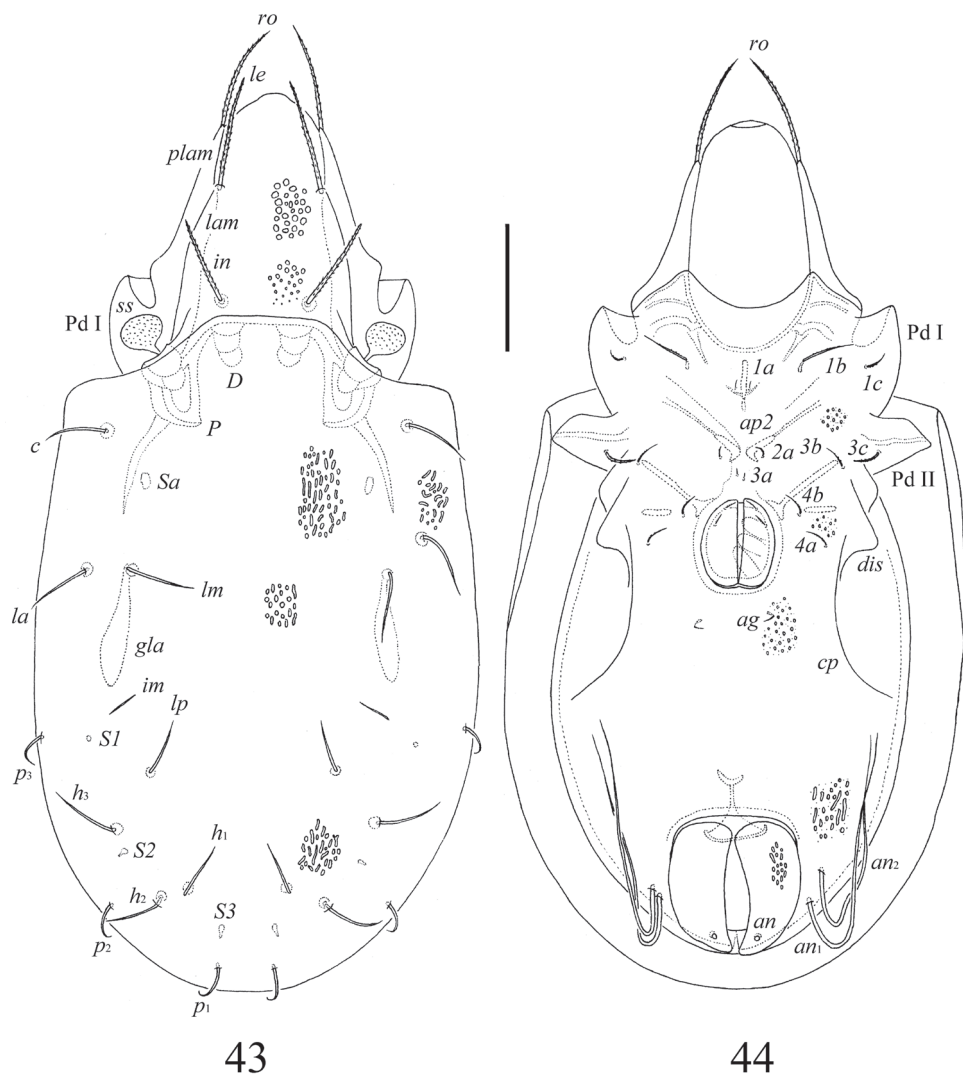
Leg	Trochanter	Femur	Genu	Tibia	Tarsus
I	v'	$d, (l), bv'', v''$	$(l), \sigma$	$(l), (v), \varphi_1, \varphi_2$	$(ft), (tc), (it), (p), (u), (a), s, (pv), e, \omega_1, \omega_2$
II	v'	$d, (l), bv'', v''$	$(l), \sigma$	$(l), (v), \varphi$	$(ft), (tc), (it), (p), (u), (a), s, \omega_1, \omega_2$
III	l', v'	d, l', ev'	l', σ	$l', (v), \varphi$	$(ft), (tc), (it), (p), (u), (a), s$
IV	v'	d, ev'	d, l'	$l', (v), \varphi$	$ft'', (tc), (p), (u), (a), s, pv''$

Roman letters refer to normal setae (ε to famulus), Greek letters to solenidia. Single prime ($'$) marks setae on anterior and double prime ($''$) setae on posterior side of the given leg segment. Parentheses refer to a pair of setae.

Benoibates minimus Mahunka, 1985

Figs 43–83

Diagnosis. Body size: 344–481 $\times$ 176–249. Body surface heavily foveolate. Rostral, lamellar and interlamellar setae setiform, barbed; latter are longest. Bothridial setae short, clavate. Ten pairs of notogastral setae of medium size. Sacculi Sa large than other. Subcapitular setae h longer than a and m . Apodemes 2 connected medially and

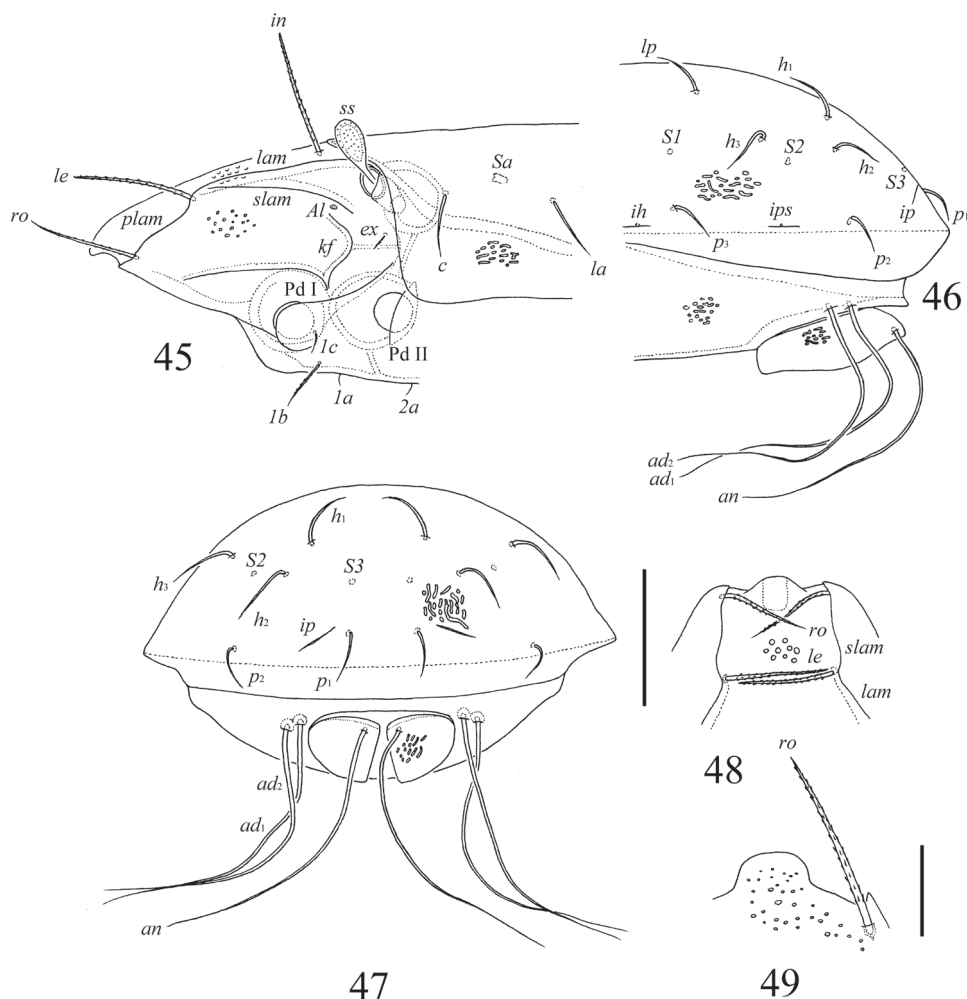


Figures 43–44. *Benoitabates minimus* Mahunka, 1985, Costa Rican specimen: **43** dorsal view **44** ventral view (gnathosoma and legs not illustrated). Scale bar 50 μ m.

removed from the anterior margin of genital aperture. Pedotecta II with one pointed tip anteriorly. Genital and aggenital setae short. Anal and adanal setae very long, flagellate.

Description. *Measurements.* Body length: 344–481 (five specimens); notogaster width: 176–249 (five specimens).

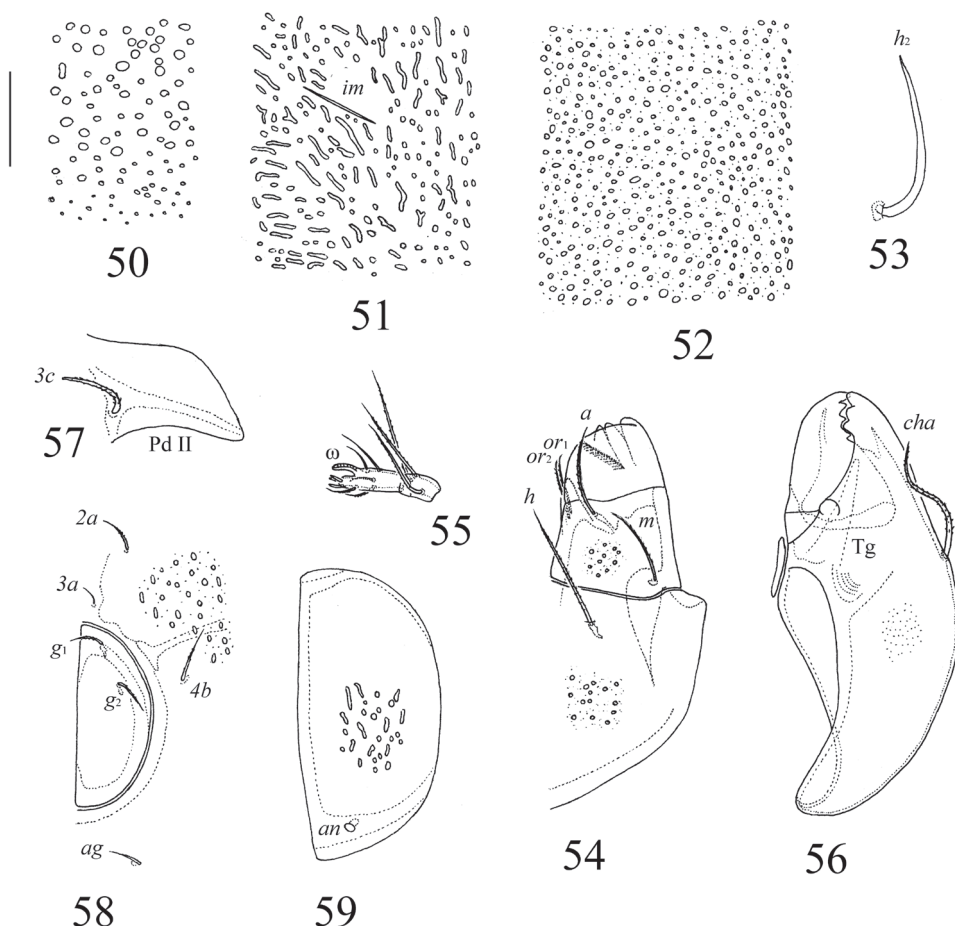
Integument. Body color yellowish brown. Body surface heavily foveolate: prodorsum with distinct, round foveoles, larger in antero-medial part (up to 4) than in basal part (up to 1); epimeral region, subcapitular mentum and gena, and genital



Figures 45–49. *Benoibates minimus* Mahunka, 1985, Costa Rican specimen: **45** lateral view of prodorsum and anterior part of notogaster and pteromorph (gnathosoma and legs I, II not illustrated) **46** lateral view of notogaster **47** posterior view of notogaster **48** frontal view of prodorsum **49** rostrum and right rostral seta in dissected specimen. Scale bars 50 μ m (45–48), 20 μ m (49).

plates with round foveoles (up to 4); notogaster and anogenital region and legs with distinct (except weak between genital and anal apertures), round or oval foveoles (up to 4), simultaneously also with longitudinal foveoles (length up to 12). Body surface of ventral side covered by microgranular cerotegument (less than 1; visible only high magnification, $\times 1000$).

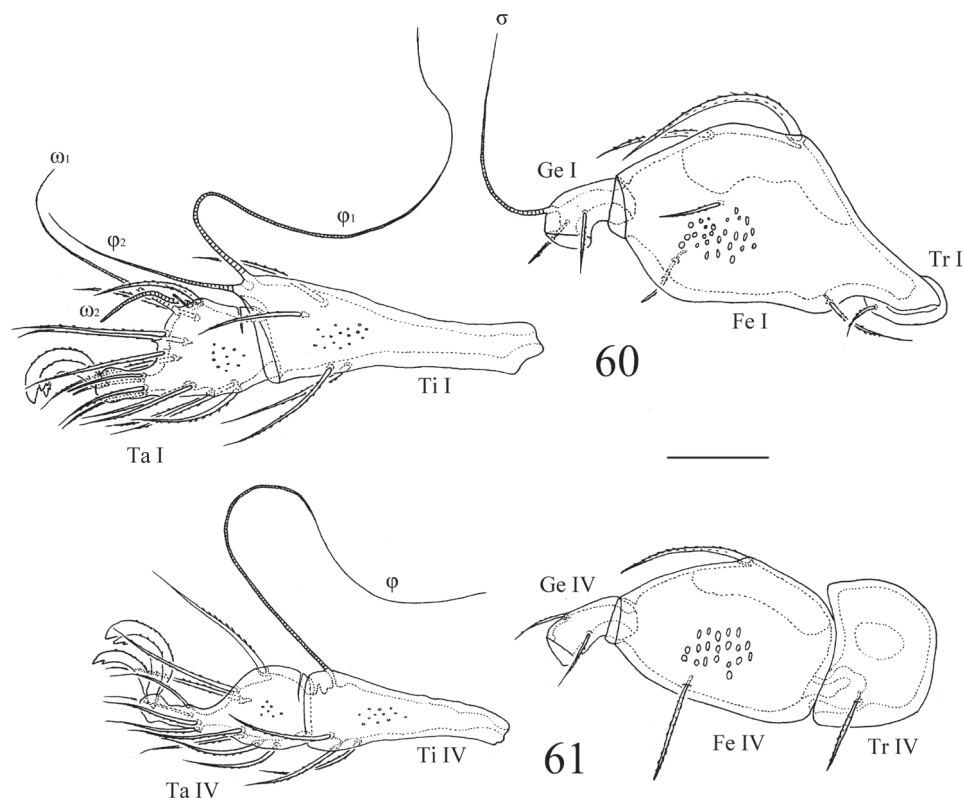
Prodorsum. Rostrum weakly protruding, rounded. Lamellae located dorso-laterally, half length of prodorsum (measured in lateral view), without cusps. Translamella absent. Prolamellar lines thin, reaching the insertions of rostral setae and bend ven-



Figures 50–59. *Benoibates minimus* Mahunka, 1985, dissected Costa Rican specimen: **50** foveoles in medio-basal part of prodorsum **51** foveoles in lateral part of notogaster and lyrifissures *im* **52** foveoles in central part of anogenital region **53** notogastral seta h_2 **54** left part of subcapitulum, ventral view **55** tarsus and tibia of palp **56** chelicera **57** pedotectum II and epimeral seta *3c* **58** left genital plate and epimeral setae *2a*, *3a*, *4b* **59** left anal plate. Scale bar 20 μ m.

trally to meet the rostral margins. Sublamellar lines distinct, long. Sublamellar porose areas small, rounded (4). Keel-shaped ridges well developed. Rostral (36–49), lamellar (41–53) and interlamellar (49–61) setae setiform, barbed. Lamellar and interlamellar straight, blunt-ended. Bothridial setae (24–32) with short stalk (8–12) and larger, clavate, barbed head (16–20). Exobothridial setae (6–8) thin, smooth.

Notogaster. Anterior notogastral margin convex, trapezoid. Dorsophragmata elongated, not reaching pleurophragmata. Notogastral shoulders rectangular in dorsal view, anterior margin almost transverse straight. Ten pairs of notogastral setae of medium size (24–36; p_1 – p_3 shorter, 20–24), setiform, smooth. Four pairs of sacculi developed: *Sa* largest, located postero-medially to setae *c*; *S1* – postero-laterally to lyrifissures *im*;

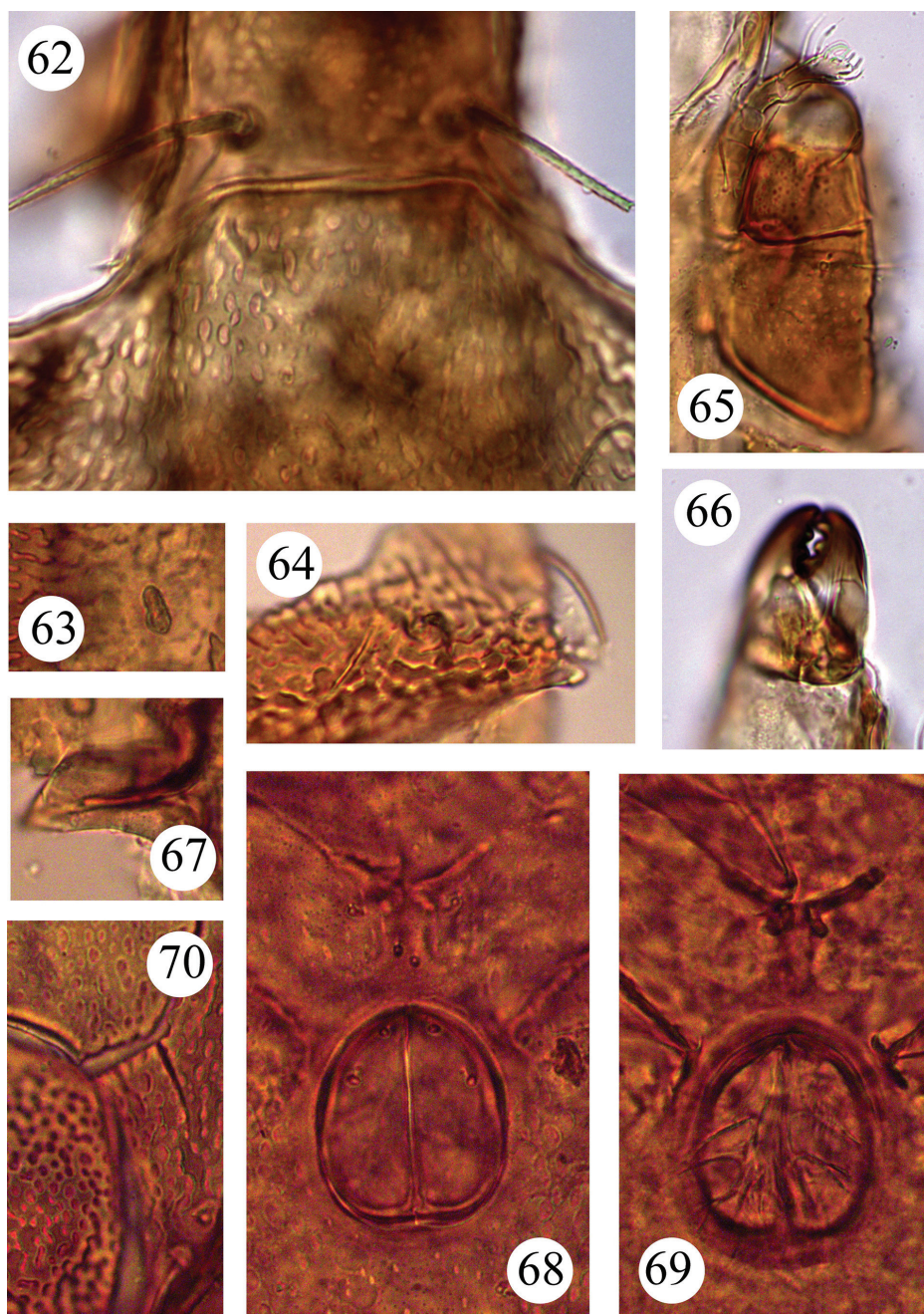


Figures 60–61. *Benoibates minimus* Mahunka, 1985, dissected Costa Rican specimen: **60** left leg I, antiaxial view **61** right leg IV, antiaxial view. Scale bar 20 μ m.

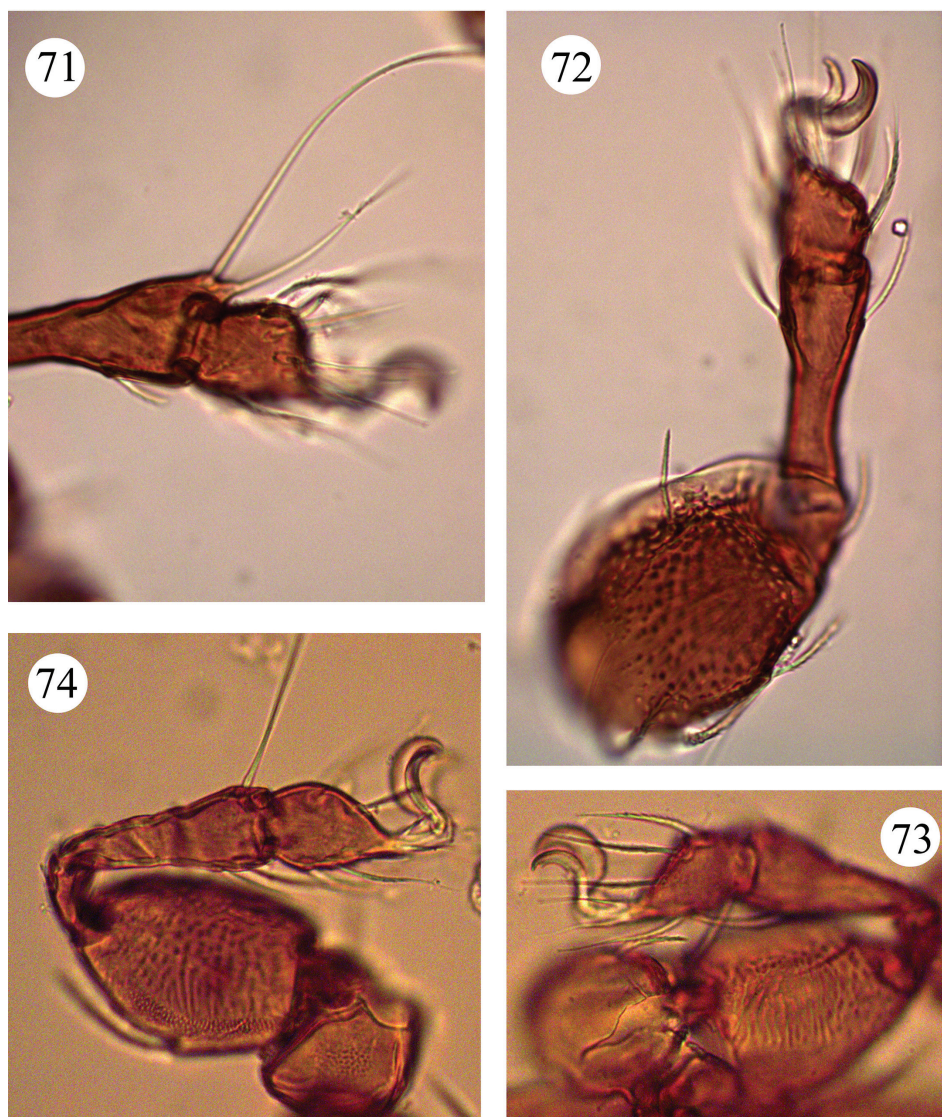
S2 – between setae h_2 and h_3 ; S3 – anteriorly to p_1 . Lyrifissures *ia* not visible; *im* located between setae *lm* and *lp*, in diagonal position; *ip* – laterally to p_1 ; *ih* – anteriorly to p_1 ; *ips* – between p_2 and p_3 . Opisthonotal glands located between setae *lm* and *lp*, but their openings not visible.

Gnathosoma. Subcapitulum longer than wide (86–98 $\times$ 61–69). Subcapitular setae setiform, slightly barbed; *h* (28–32) slightly thicker and longer than *a* and *m* (both 18–20). Two pairs of adoral setae (12) setiform, densely barbed. Palps (length 45–53) with setation 0–2–1–3–9(+ ω). Solenidion attached to eupathidium. Chelicerae (length 90–102) with one barbed setae (*cha*, 28–36), *chb* and their alveoli absent. Trägårdh's organ long, tapered.

Epimeral and lateral podosomal regions. Apodemes 1, 2, 3, sejugal and sternal apodemes distinct. Apodemes 2 connected medially and removed from the anterior margin of genital aperture. Sternal apodeme of medium size, not reaching the apodemes 2. Epimeral setal formula: 3–1–3–2. Centrovventral setae *1a*, *3a* smooth, other slightly barbed; *1b* (20) longer than *1c*, *2a*, *3b*, *3c*, *4a*, *4b* (12–16) and *1a*, *3a* (4–6). Setae *3c* thickest. Pedotecta I large, concave (measured in dorsal view) and scale-like (measured



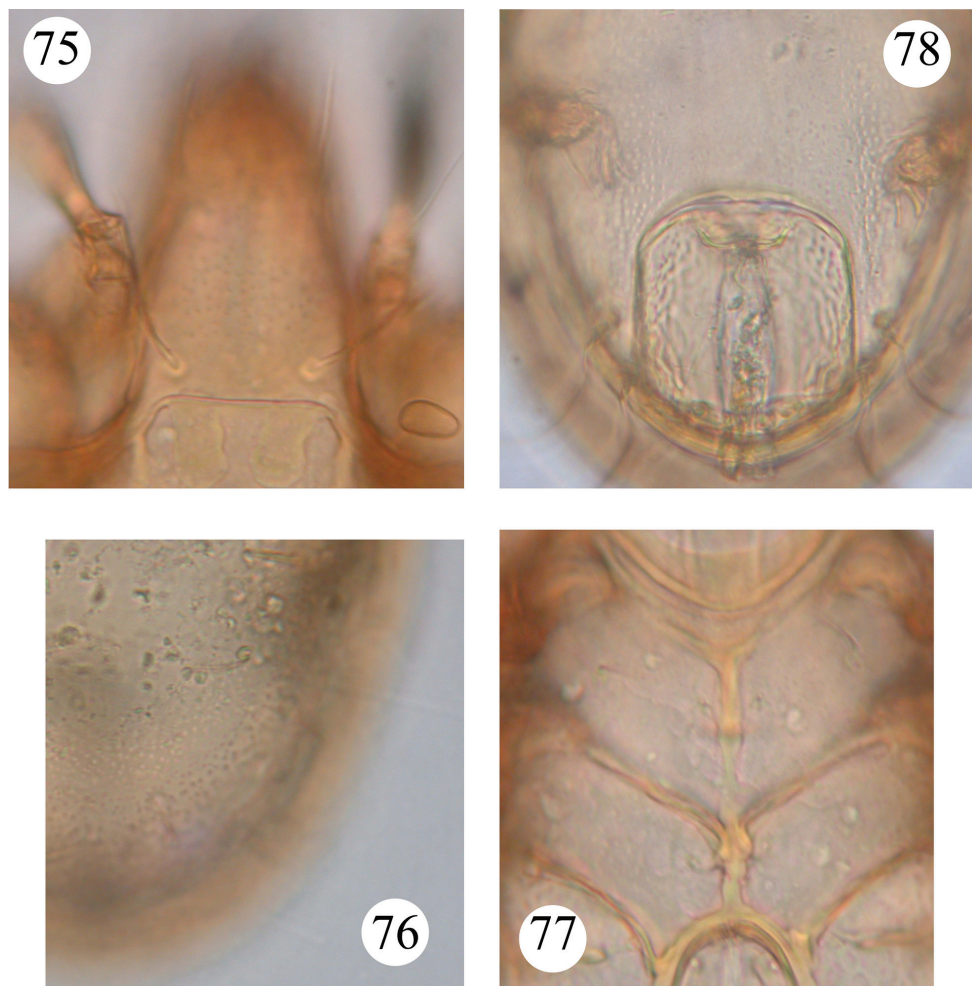
Figures 62–70. *Benoitibates minimus* Mahunka, 1985, dissected Costa Rican specimen, microscope images: **62** dorsal view of basal part of prodorsum and anterior notogastral margin **63** saccule *Sa* **64** lyrifissure *ip* and foveoles in posterior part of notogaster **65** right part of subcapitulum, ventral view, and right palp **66** antero-medial part of chelicera **67** ventral view of right pedotectum II **68** genital plates and central part of epimeral region **69** genital plates and central part of epimeral region **70** medio-lateral part of left anal plate, insertions of adanal setae, and foveoles in anogenital region. Scale bar 50 μ m.



Figures 71–74. *Benoitibates minimus* Mahunka, 1985, dissected Costa Rican specimen, microscope images: **71** tarsus and antero-medial part of tibia of leg I, left, paraxial view **72** leg II, left, antiaxial view **73** leg III, right, antiaxial view **74** leg IV, left, paraxial view. Scale bar 50 μ m.

in lateral view); pedotecta II smaller, trapezoid, with one pointed tip anteriorly (measured in ventral view) and scale-like (measured in lateral view). Discidia elongated, weakly triangular. Circumpedal carinae distinct.

Anogenital region. Two pairs of genital setae (8) setae thin, slightly barbed. One pair of aggenital setae (6–8) setae thin, smooth. One pair of anal (106–135) and two pairs of adanal setae (114–143) very long, flagellate. Often anal setae brokened, only alveoli visible. Lyrifissures *iad* not visible.

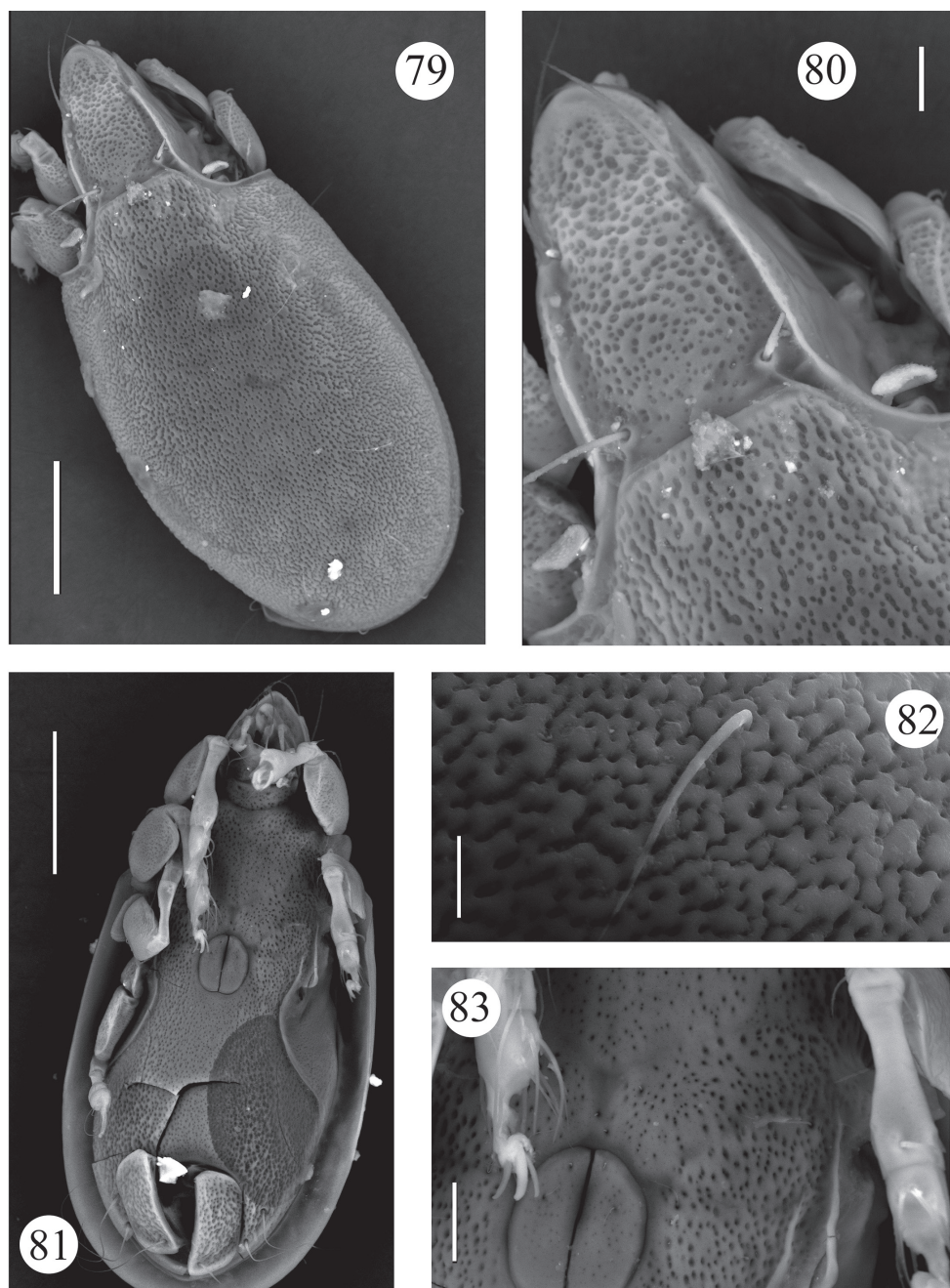


Figures 75–78. *Benoibates minimus* Mahunka, 1985, holotype, microscope images: **75** dorsal view of prodorsum and anterior part of notogaster **76** dorso-lateral part of notogaster **77** epimeral region **78** ano-adanal region.

Legs. Median claw weakly thicker than two lateral claws; all with several minute barbs on dorsal side. Lateral claws with ventral tooth. Formulae of leg setation and solenidia: I (1–5–2–4–16) [1–2–2], II (1–5–2–4–13) [1–1–2], III (2–3–1–3–13) [1–1–0], IV (1–2–2–3–11) [0–1–0]; homology of setae and solenidia indicated in Table 1. Famulus short, straight, slightly dilated distally, truncated. Solenidia ω_2 on tarsi I, ω_1 and ω_2 on tarsi II, σ on genua II, III of medium size, thickened, blunt-ended. Other solenidia long, setiform.

Remarks. Costa Rican specimens of *Benoibates minimus* are similar in all morphological characters to Antilles specimens from the original description (Mahunka 1985).

Distribution. Neotropical region.



Figures 79–83. *Benoibates minimus* Mahunka, 1985, Costa Rican specimen, SEM micrographs: **79** dorsal view **80** dorsal view of prodorsum and anterior part of notogaster **81** ventral view **82** notogastral seta h_2 **83** genital plates and left part of epimeral region. Scale bar 100 μm (**79**, **81**), 20 μm (**80**, **83**), 10 μm (**82**).

Key to known species of *Benoibates*¹

- 1 Anal and adanal setae of medium size, similar to length of anal plate or shorter **2**
- Anal and adanal setae long, flagellate, longer than length of anal plate **4**
- 2 Anal and adanal setae setiform, twice shorter than length of anal plate; body size: 374–600 × 200–330 ***B. juglans* (Jacot, 1938)**. U.S.A.
- Anal and adanal setae flagellate, similar in length to anal plate **3**
- 3 Interlamellar setae dilated distally; body size: 475 × 221 ***B. amazonicus* Balogh & Mahunka, 1969(b)**. Neotropical region
- Interlamellar setae setiform; body size: 358–625 × 165–275 ***B. muscicola* Baranek, 1981**. Argentina
- 4 Lamellar setae with rounded tip, interlamellar setae dilated distally; body size: 380–472 × 255–270 ***B. flagellifer* Balogh, 1958 (see also Balogh 1960)**. Angola
- Lamellar and interlamellar setae simple, setiform **5**
- 5 Interlamellar setae very long, reaching the rostrum; body size: 448–584 × 210–290 ***B. borhidii* Balogh & Mahunka, 1980**. Neotropical region
- Interlamellar setae of medium size, not reaching the rostrum **6**
- 6 Notogaster with 11 pairs of setae (c_1 present); body size: 665–680 × 339–388... ***B. plurisetus* Mahunka, 1984**. Paraguay
- Notogaster with 10 pairs of setae (c_1 absent) **7**
- 7 Adanal region with foveoles, forming the longitudinal lineate rows **8**
- Adanal region with foveoles, not forming the longitudinal lineate rows **9**
- 8 Translamella present; epimeral setae $1b$ considerable longer than $1c$; body size: 355–480 × 185–272 ***B. rugosus* Mahunka, 2001**. Kenya
- Translamella absent; epimeral setae $1b$ and $1c$ similar in length; body length: 375 ***B. marginatus* (Hammer, 1973)**. Polynesia
- 9 Apodemes 2 connected to anterior margin of genital aperture; lyrifissures im in transverse position; body size: 514–597 × 265–332 ***B. bolivianus* Balogh & Mahunka, 1969(a)**. Neotropical region
- Apodemes 2 not connected to anterior margin of genital aperture; lyrifissures im in diagonal position **10**
- 10 Distance ad_1 – ad_1 equal to ad_2 – ad_2 ; bothridial head small; body size: 585–647 × 369–388 ***B. chacoensis* Mahunka, 1984**. Paraguay
- Distance ad_1 – ad_1 smaller than ad_2 – ad_2 ; bothridial head large; body size: 344–481 × 176–249 ***B. minimus* Mahunka, 1985**. Neotropical region

¹ *Benoibates crinitus* (Berlese, 1910) was very poorly described by Berlese (1910), therefore we did not include this species in the key.

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Three new species of the *Stenus cirrus* group (Coleoptera, Staphylinidae) from Jiangxi, South China

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Abstract

Three new species from Jiangxi, China, are described and illustrated: *S. wugongshanus* **sp. n.**, *S. mingyueshanus* **sp. n.**, and *S. songxiaobini* **sp. n.** A previously published key to the Chinese species of the *Stenus cirrus* group is modified to accommodate the new species.

Keywords

Coleoptera, Staphylinidae, *Stenus*, *cirrus* group, China, new species, key to species

Introduction

The speciose *Stenus cirrus* group currently holds 63 species worldwide, 46 of them are known to occur in China and two in Jiangxi. Members of the group are characterized by the presence of long and erect setae on the abdomen. A detailed definition of the group was given in Tang et al. (2008) and Puthz (2009).

Among the specimens we recently collected in the Luoxiao Mountains, Jiangxi Province, three species are recognized as new and are described herein.

Material and methods

For examination of the male genitalia, the apical three abdominal segments were detached from the body after softening in hot water. The aedeagi, together with other dissected parts, were mounted in Euparal (Chroma Gesellschaft Schmidt, Koengen, Germany) on plastic slides. Photos of sexual characters were taken with a Canon G9 camera attached to an Olympus CX31 microscope; habitus photos were taken with a Canon macro photo lens MP-E 65 mm attached to a Canon EOS7D camera.

The type specimens treated in this study are deposited in the following collections:

SHNU Department of Biology, Shanghai Normal University, P. R. China

cPut Private collection Volker Puthz, Schlitz, Germany

The measurements are abbreviated as follows:

BL	body length, measured from the anterior margin of the clypeus to the posterior margin of abdominal tergite X
FL	forebody length, measured from the anterior margin of the clypeus to the apicolateral angle of elytra
HW	width of head including eyes
PW	width of pronotum
EW	width of elytra
PL	length of pronotum
EL	length of elytra, measured from humeral angle
SL	length of elytral suture

Taxonomy

Stenus wugongshanus sp. n.

<http://zoobank.org/5FFA59B8-571C-472E-A91C-66B24552A881>

Figs 1, 2, 7–17

Type material. Holotype: CHINA: Jiangxi Prov.: ♂, Pingxiang City, Wugong Shan National Park, alt. 1500–1750 m, 21.VII.2013, Song, Yin & Yu leg. (SHNU). **Paratypes:** CHINA: Jiangxi Prov.: 1♂, 4♀♀, same data as holotype (SHNU); 6♂♂, 3♀♀, same locality, but alt. 1000–1350 m, 20.VII.2013, Song, Yin & Yu leg. (1♂, 1♀ in cPut, rest in SHNU).

Description. BL: 2.9–3.4 mm, FL: 1.4–1.7 mm.

HW: 0.66–0.77 mm, PL: 0.47–0.54 mm, PW: 0.48–0.56 mm, EL: 0.49–0.51 mm, EW: 0.54–0.63 mm, SL: 0.35–0.40 mm.



Figures 1, 2. Habitus of *Stenus wugongshanus* in dorsal and ventral view. Scale bar: 1 mm.

Brachypterous. Head blackish, body reddish brown, each elytron with a large ill-defined orange spot, which is about $2/3$ as long and about $1/2$ as broad as the respective elytron, abdomen shiny; antennae reddish yellow, club infusate; maxillary palpi and legs reddish yellow; clypeus brown, labrum reddish brown; moderately sparsely pubescent. Paraglossae oval.

Head 1.17–1.22 times as wide as elytra; interocular area with two deep longitudinal furrows, median portion slightly convex, not reaching level of inner eye margins, with a broad impunctate line along midline; punctures round, slightly larger in median

portion than near inner margins of eyes, diameter of large punctures about as wide as apex of 2nd antennal segment in cross section, interstices between punctures faintly microsculptured, narrower than half the diameter of punctures. Antennae, when reflexed, slightly extending beyond posterior margin of pronotum, antennomeres III–VIII distinctly narrower than II; IX–XI gradually broadened, forming loose club; relative length of segments from base to apex as 10.5: 6.5: 13.0: 7.5: 7.0: 5.0: 5.5: 3.5: 5.0: 6.0: 6.5.

Pronotum 0.95–1.00 times as long as wide, widest near middle and constricted at base; disk uneven, with distinct median longitudinal furrow of about half the length of pronotum and with an impunctate line along the middle; punctures round and moderately confluent, variable in size, diameter of large punctures about as wide as middle of 2nd antennal segment in cross section, interstices with dense microsculpture, much narrower than half the diameter of punctures.

Elytra 0.92–0.97 times as long as wide, distinctly constricted at base, lateral margins gradually divergent posteriad; disk uneven with distinct humeral impression, distinct sutural impression and rather faint postero-lateral impression; punctures fusiform and confluent, larger than those on pronotum, interstices between punctures microsculptured, much narrower than half the diameter of punctures.

Hind tarsi 0.70–0.75 times as long as hind tibiae, tarsomeres IV strongly bilobed.

Abdomen cylindrical; paratergites very narrow and smooth, present only in segment III, segments IV–VI with tergites and sternites entirely fused and traces of paratergites present only at base of each segment, posterior margin of tergite VII with indistinct palisade fringe; punctures round, dense at the base of each tergite, gradually becoming smaller posteriad, interstices smooth.

Male. Sternite VII with shallow emargination at middle of posterior margin, sternite VIII (Fig. 7) with semi-circular emargination at middle of posterior margin; sternite IX (Fig. 8) with long apicolateral projections, posterior margin serrate and slightly produced in the middle; tergite X (Fig. 9) with posterior margin truncate and slightly emarginated in the middle. Aedeagus (Figs 10, 11) with median lobe subparallel-sided, apical sclerotized portion triangular with round projection at apex, expulsion hooks (Fig. 13) large, strongly sclerotized; parameres slender and almost straight, extending a little beyond apex median lobe, swollen at apex, each with about 11 setae on apico-internal margin (Fig. 12).

Female. Abdomen slightly broader than that of male; sternite VIII (Fig. 14) with posterior margin weakly pointed at middle; tergite X (Fig. 15) with posterior margin truncate; spermathecal duct with basal portion strongly sclerotized and the remainder weakly sclerotized (Figs 16, 17).

Distribution. Jiangxi Province: Wugong Shan (China).

Diagnoses. *Stenus wugongshanus* resembles *S. huangganmontium* Puthz, 2003 (Wuyishan, Jiangxi Province) in most aspects, but may be distinguished by the lack of paratergites on segments IV and V, which are present in *S. huangganmontium*.

Etymology. The specific name is derived from “Wugongshan”, the type locality of this species.

***Stenus mingyueshanus* sp. n.**

<http://zoobank.org/2C320675-82A8-425E-A2D7-18490431CE6F>

Figs 3, 4, 18–27

Type material. Holotype: CHINA: Jiangxi Prov.: ♂, Yichun City, Mingyue Shan, alt. 1140 m, 23.X.2013, Peng, Shen & Yan leg. (SHNU). **Paratypes:** CHINA: Jiangxi Prov.: 9♂♂, 5♀♀, same data as holotype (1♂, 1♀ cPut, rest in SHNU); 2♂♂, 1♀, same data but alt. 1600 m, 22.X.2013 (SHNU); 1♂, 2♀♀, Yichun City, Mingyueshan National Park, alt. 1610 m, 11.VII.2013, Song, Yin & Yu leg. (SHNU).

Description. BL: 4.2–5.1 mm, FL: 2.0–2.3 mm.

HW: 0.83–0.91 mm, PL: 0.65–0.72 mm, PW: 0.63–0.69 mm, EL: 0.74–0.82 mm, EW: 0.74–0.84 mm, SL: 0.53–0.59 mm.

Brachypterous. Body blackish, each elytron with an orange marking near lateral margin, this marking about 2/3 as long and about 1/2 as broad as the respective elytron; antennae reddish yellow, club infusate; maxillary palpi and legs reddish yellow; clypeus black, labrum brown, moderately densely pubescent. Paraglossae oval.

Head 1.08–1.12 times as wide as elytra; interocular area with two deep longitudinal furrows, median portion convex, not reaching level of inner eye margins, with a broad impunctate line along midline; punctures round to fusiform, larger and sparser in median area than near inner margins of eyes; diameter of large punctures about as wide as middle of 2nd antennal segment in cross section, interstices smooth, varying from narrower to slightly broader than half the diameter of punctures. Antennae, when reflexed, slightly extending beyond posterior margin of pronotum, antennomeres III–VIII segments distinctly narrower than II; IX–XI gradually broadened, forming loose club; relative length of segments from base to apex as 11.5: 7.5: 20.0: 10.5: 10.5: 8.5: 7.0: 5.0: 6.5: 6.0: 8.0.

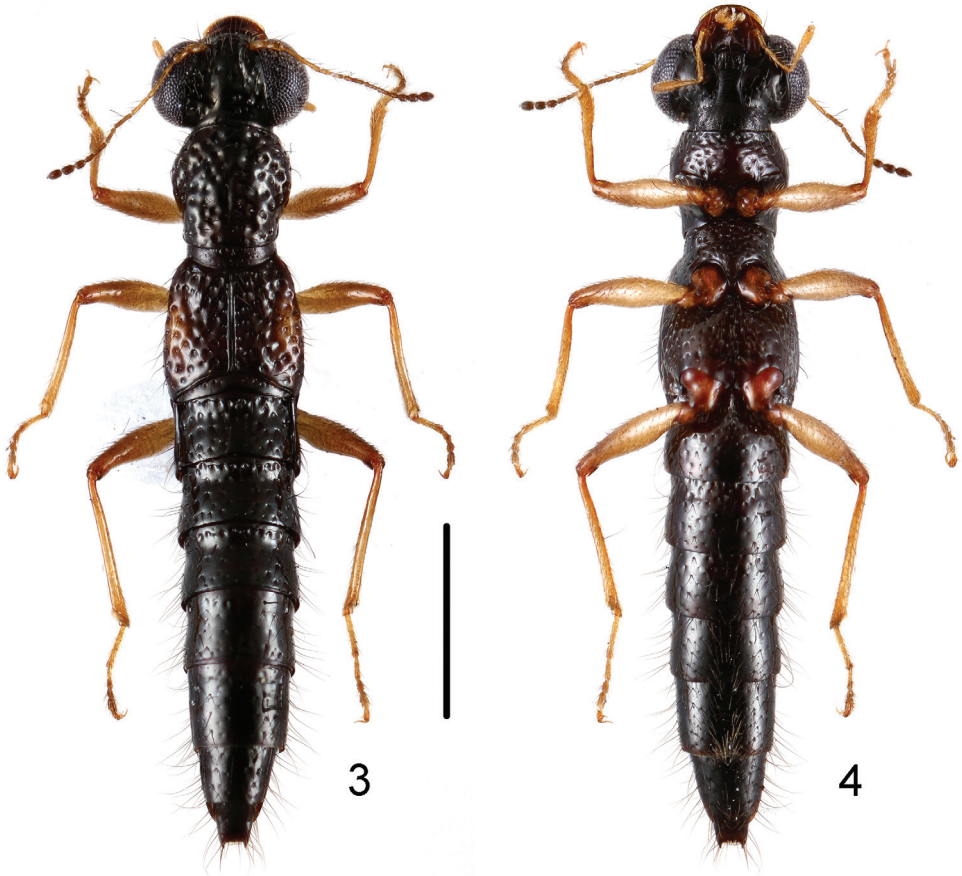
Pronotum 1.01–1.04 times as long as wide, widest a little before middle and constricted at base; with shallow median longitudinal furrow of about half the length of pronotum; punctures round, some of them confluent, variable in size, diameter of large punctures much larger than middle of 2nd antennal segment in cross section, interstices smooth, distinctly narrower than half the diameter of punctures.

Elytra 0.92–1.02 times as long as wide; distinctly constricted at base, lateral margins gradually divergent posteriad; disk uneven with distinct humeral impression and faint sutural impression; punctures similar to those of pronotum, interstices smooth, narrower than half the diameter of punctures.

Hind tarsi 0.72–0.74 times as long as hind tibiae, tarsomeres IV strongly bilobed.

Abdomen cylindrical; paratergites very narrow and punctate, present only in segment III, segments IV–VI with tergites and sternites entirely fused and traces of paratergites present only at base of each segment, posterior margin of tergite VII with indistinct palisade fringe; punctures round, gradually becoming smaller posteriad, interstices smooth, varying from narrower to broader than diameter of punctures.

Male. Sternite VII with shallow emargination in the middle of posterior margin, sternite VIII (Fig. 18) with triangular emargination in the middle of posterior margin;



Figures 3, 4. Habitus of *Stenus mingyueshanus* in dorsal and ventral view. Scale bar: 1 mm.

sternite IX (Fig. 19) with long and acute apicolateral projections, posterior margin serrate and nearly straight; tergite X (Fig. 20) with posterior margin convex. Aedeagus (Figs 21, 22) with median lobe subparallel-sided in basal portion and tapering in apical half, apical sclerotized portion nearly triangular, expulsion hooks (Fig. 24) large, strongly sclerotized; parameres almost straight, distinctly longer than median lobe, each with about 12–13 setae on apico-internal margin (Fig. 23).

Female. Sternite VIII (Fig. 25) with posterior margin rounded; tergite X (Fig. 26) slightly emarginated at middle of posterior margin; without sclerotized spermatheca (Fig. 27).

Distribution. Jiangxi Province: Mingyue Shan (China).

Diagnoses. *Stenus mingyueshanus* resembles *S. ovalis* Tang, Li & Zhao, 2005 (Wuyanling, Zhejiang Province), but may be distinguished by the distinctly coarser and sparser punctation of the forebody in the new species.

Etymology. The specific name is derived from “Mingyueshan”, the type locality of this species.



Figures 5, 6. Habitus of *Stenus songxiaobini* in dorsal and ventral view. Scale bar: 1 mm.

***Stenus songxiaobini* sp. n.**

<http://zoobank.org/4B067492-9B22-4426-8B8E-99D7956193CD>

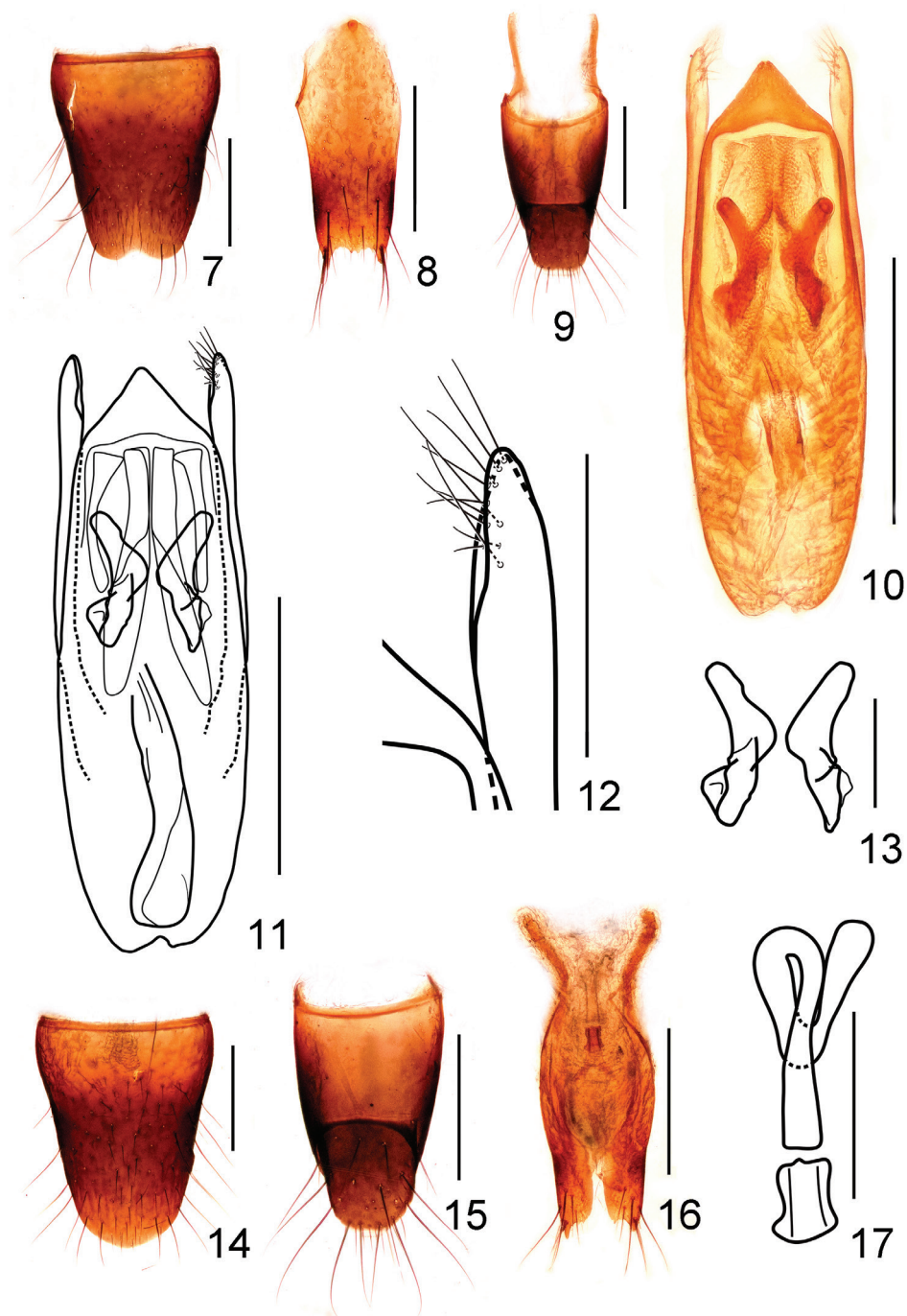
Figs 5, 6, 28–37

Type material. **Holotype:** CHINA: Jiangxi Prov.: ♂, Pingxiang City, Wugong Shan National Park, alt. 1340–1400 m, 19.VII. 2013, Song, Yin & Yu leg. (SHNU). **Paratypes:** CHINA: Jiangxi Prov.: 1♂, 1♀, same data, but alt. 1000–1350 m, 20.VII.2013 (SHNU).

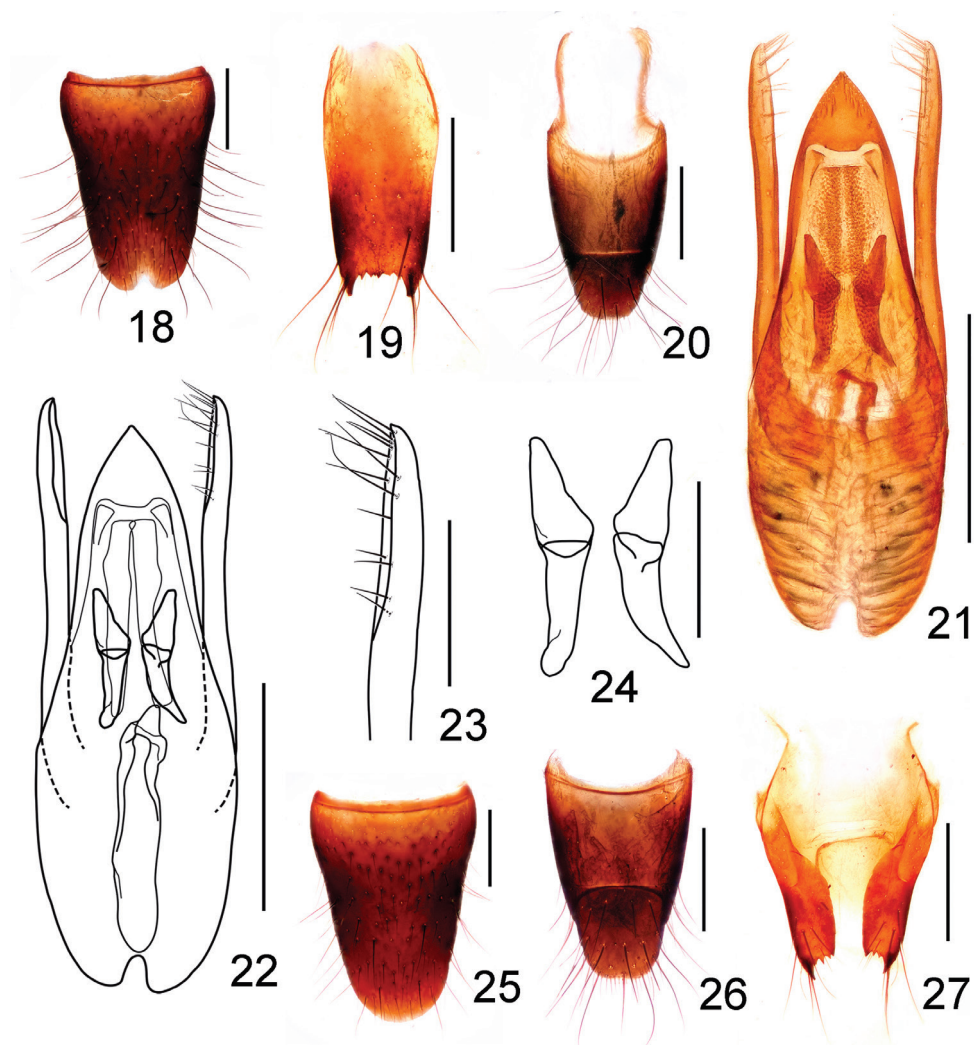
Description. BL: 4.3–4.7 mm, FL: 2.0–2.1 mm.

HW: 0.82–0.83 mm, PL: 0.62–0.67 mm, PW: 0.63–0.64 mm, EL: 0.74–0.78 mm, EW: 0.73–0.76 mm, SL: 0.52–0.57 mm.

The new species is similar to *S. mingyueshanus* sp. n. in most respects, but different in the following characters: head 1.08–1.12 times as wide as elytra; frons with interstices smooth, varying from narrower to slightly broader than half the diameter



Figures 7–17. *Stenus wugongshanus*. **7** male sternite VIII **8** male sternite IX **9** male tergites IX, X **10**, **11** aedeagus **12** apical portion of paramere **13** expulsion hooks **14** female sternite VIII **15** female tergites IX, X **16** valvifers and spermatheca **17** spermatheca. Scale bars: **12–13**: 0.1 mm; **7–11**, **14–17**: 0.25 mm.

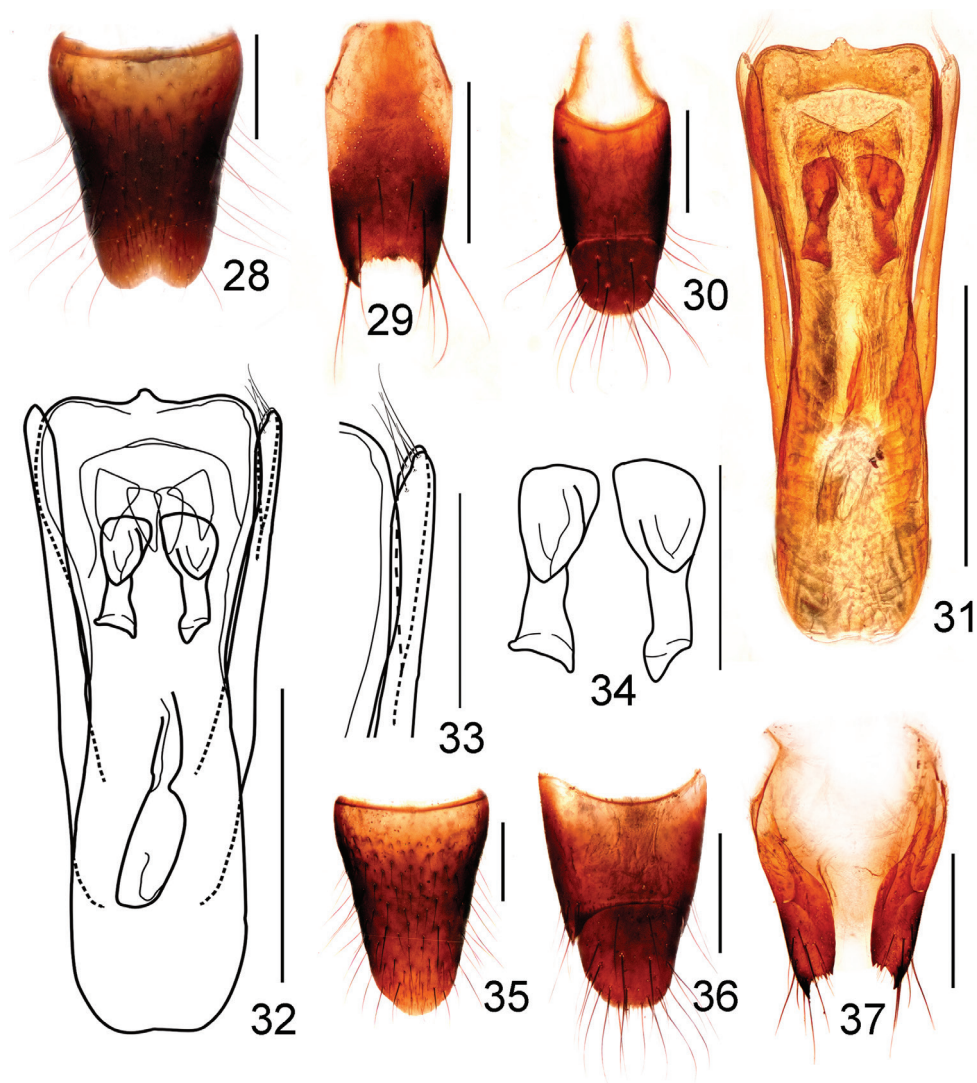


Figures 18–27. *Stenus mingyueshanus*. **18** male sternite VIII **19** male sternite IX **20** male tergites IX, X **21, 22** aedeagus **23** apical portion of paramere **24** expulsion hooks **25** female sternite VIII **26** female tergites IX, X **27** valvifers. Scale bars: **23–24**: 0.1 mm; **18–22, 25–27**: 0.25 mm.

of punctures; relative length of antennal segments from base to apex as 9.5: 7.0: 18.5: 10.5: 8.5: 8.0: 6.5: 4.0: 6.0: 6.0: 7.0.

Pronotum 1.08–1.13 times as long as wide; median longitudinal furrow shallow of about $\frac{2}{5}$ the length of pronotum, punctures round and partly confluent, variable in size, diameter of large punctures slightly larger than middle of 2nd antennal segment in cross section; interstices smooth, distinctly narrower than half the diameter of punctures.

Elytra 1.02–1.03 times as long as wide; size of punctures similar to those of pronotum, interstices partly with faint reticulation, somewhat narrower than half the diameter of punctures.



Figures 28–37. *Stenus songxiaobini*. **28** male sternite VIII **29** male sternite IX **30** male tergites IX, X **31, 32** aedeagus **33** apical portion of paramere **34** expulsion hooks **35** female sternite VIII **36** female tergites IX, X **37** valvifers. Scale bars: **33–34**: 0.1 mm; **28–32, 35–37**: 0.25 mm.

Hind tarsi 0.77–0.80 times as long as hind tibiae.

Male. Sternite VII with shallow emargination in the middle of posterior margin, sternite VIII (Fig. 28) with a circular emargination in the middle of posterior margin; sternite IX (Fig. 29) with long and acute apicolateral projections, posterior margin serrate and nearly straight; tergite X (Fig. 30) with posterior margin broadly rounded. Aedeagus (Figs 31, 32) with median lobe subparallel-sided in basal portion and broadening in apical half, apical margin truncate with a median projection, expulsion hooks

(Fig. 34) large, strongly sclerotized; parameres almost straight, extending a little beyond apex of median lobe, each with about 5 setae on apico-internal margin (Fig. 33).

Female. Sternite VIII (Fig. 35) with convex posterior margin; tergite X (Fig. 36) with posterior margin convex; spermatheca unsclerotized (Fig. 37).

Distribution. Jiangxi Province: Wugong Shan (China).

Diagnoses. *Stenus songxiaobini* is similar to *S. mingyueshanus* sp. n., but may be distinguished from the latter by denser and deeper punctures on the pronotum and elytra and by the shallower median longitudinal furrow, which is about 2/5 the length of pronotum.

Etymology. This species is named in honor of Mr. Xiao-Bin Song, collector of the new species.

Modified couplets of the key (Pan et al. 2012) to Chinese species of the *Stenus cirrus* group

- 10a Pronotum with interstices distinctly reticulated. Habitus: Figs 1, 2; sexual characters: Figs 7–17. BL: 2.9–3.4 mm. China: Jiangxi: Wugongshan..... *S. wugongshanus* sp. n.
- Pronotum with interstices smooth **10b**
- 10b Head narrower than elytra or nearly as wide as elytra **11**
- Head distinctly wider than elytra **13**
- 18a Larger species, FL $\geq$ 1.9 mm; elytral markings longer than half the length of elytra, extending towards humeral angles **18b**
- Smaller species, FL = 1.6–1.7 mm, elytral markings shorter than half the length of elytra, not extending towards humeral angles. Habitus: figure 1 in Tang et al. (2005); sexual characters: figures 4–7 in Tang et al. (2005). BL: 3.2–4.1 mm. China: Hubei: Houhe *S. andoi* Tang, Li & Zhao, 2005
- 18b Punctures on pronotum very large, diameter of large punctures much larger than middle of 2nd antennal segment in cross section. Habitus: Figs 3–4; sexual characters: 18–27. BL: 4.2–5.1 mm. China: Jiangxi: Mingyueshan *S. mingyueshanus* sp. n.
- Punctures on pronotum relatively small, diameter of large punctures slightly larger than middle of 2nd antennal segment in cross section..... **18c**
- 18c Pronotum without median longitudinal furrow; punctuation of abdominal tergites coarser, punctures on abdominal tergite IV as large as those of elytra. Habitus: figure 3 in Tang, Li & Zhao (2005); sexual characters: figures 12–15 in Tang et al. (2005). BL: 3.8–4.5 mm. China: Zhejiang: Wuyanling *S. ovalis* Tang, Li & Zhao, 2005
- Pronotum with shallow median longitudinal furrow; punctuation of abdominal tergites less coarse, punctures on abdominal tergite IV smaller than those of elytra. Habitus: Figs 5, 6; sexual characters: Figs 28–37. BL: 4.3–4.7 mm. China: Jiangxi: Wugongshan *S. songxiaobini* sp. n.

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We would like to express our sincere gratitude to Dr. V. Puthz (Germany) for his constant guidance on our study of Chinese Steninae and to all the collectors mentioned in the paper. The study is supported by the National Natural Science Foundation of China (No. 31101659, No. 31201734) and Shanghai Normal University (DZL125).

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***Cryptalyra* (Hymenoptera, Megalyridae) from Maranhão, Brazil: three new species discovered after a large collecting effort**

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Abstract

Cryptalyra is a small genus of megalyrid wasps with three described species confined to South America. Our main goal in this work is to record an increase in the known diversity, adding the three new species *Cryptalyra helenae* sp. n., *C. ichiroi* sp. n. and *C. limeirai* sp. n., from a single locality in Maranhão, Brazil that has been subjected to extraordinary collecting effort. We are providing a key for the species of the genus, as well as illustrations of the main structures for recognition of the new taxa.

Keywords

Apocrita, Cryptalyrini, Neotropical, taxonomy, Malaise trap

Introduction

Megalyridae have 49 extant species allocated in eight extant genera (Vilhelmsen et al. 2010). The information available comes primarily from a few members of *Megalyra* (Rodd 1951, Naumann 1987, Shaw 1990a) most attacking xylophagous Coleoptera.

Shaw (1987, 1988, 1990a) surveyed and revised the majority of extant Megalyridae; Shaw (1990b) summarized the classification, analyzed the phylogeny of the extant and extinct members of the family known at the time.

Shaw (1987) described three monotypic genera from the Neotropics: *Cryptalyra* Shaw from the tropical and subtropical part east of the Andes, and *Neodinapsis* Shaw and *Rigel* Shaw from Chile in the temperate Neotropics. In terms of species, the Neotropical megalyrid fauna seems to be very depauperate. However, the extant generic diversity is on a par with that of the other Southern Hemisphere regions, and, as in the Afrotropics, the poor species turnout might yet prove to be the result of lack of exploration.

Cryptalyra Shaw, 1987 is a small genus confined to South America (Azevedo and Tavares 2006). They are *C. colombia* Shaw, 2003, from Colombia, *C. plaumanni* Shaw, 1987 from Southern Brazil and *C. depressa* Azevedo and Tavares, 2006 from Northern Brazil. All of them were described based on a single female.

While visiting the collection of Universidade Estadual do Maranhão in Caxias we found ten more specimens of *Cryptalyra*, which correspond to three new species. Thus the main goal of this contribution is to formally describe them.

Material and methods

The material examined was provided by Francisco Limeira-de-Oliveira, curator of the Coleção Zoológica do Maranhão (CZMA), Universidade Estadual do Maranhão (UEMA), Caxias, Maranhão, Brasil.

The terminology follows Shaw (1987, 2003), Vilhelmsen et al. (2010) and the Hymenoptera Anatomy Ontology (HAO) project (Yoder et al. 2010, Seltmann et al. 2012). The nomenclature of integumental sculpture follows Harris (1979). The key to species follows Azevedo and Tavares (2006). All new species have been prospectively registered with Zoobank (Polaszek et al. 2005).

The specimens were examined under a Leica MZ80 Stereo Microscope. Images were taken with a Leica DFC 495 video camera attached to a Leica Z16 APO with a Planapo 2.0x objective. Figures were produced from stacks of images that vertically transected the specimen using Leica LAS (Leica Application Suite V4.3.0) Microsystems by Leica (Switzerland) Limited. These were combined automatically into a single image using Helicon Focus (version 6.0.18), based on Method C (Pyramid) and focus autoadjustments 1% (horizontally).

Results

Key to females of *Cryptalyra* modified from Azevedo and Tavares (2006)

- 1 Propodeum with wide longitudinal depression 2
- Propodeum without such depression (Figs 6B, 7B, 8B) 3
- 2 Body light chestnut brown; middle of face to vertex crest without longitudinal sulcus; median mesoscutal line as continuous sulcus; propodeum with longitudinal depression delimited by lateral carina..... ***C. depressa* Azevedo & Tavares**

- Body black; middle of face to vertex crest with longitudinal sulcus (Figs 4A/B); median mesoscutal line deep, broad and scrobiculate (Fig. 6B); propodeum with longitudinal depression not delimited by lateral carina..... *C. belenae* sp. n. (Fig. 1)
- 3 Propodeum with posterior tubercle..... *C. plaumanni* Shaw
- Propodeum without posterior tubercle (Figs 8, 9)..... 4
- 4 Median mesoscutal line formed by continuous fovea, unbounded and widening posteriorly (Fig. 8B); metatibia with two apical spurs (Fig. 12F)..... *C. limeirai* sp. n. (Fig. 3)
- Median mesoscutal line formed by separated fovea (Fig. 7B); metatibia with one apical spur..... 5
- 5 Eye densely setose; frons densely foveolate; hind wing Rs vein not reaching middle of wing; lower valve with three minute apical teeth..... *C. colombia* Shaw
- Eye sparsely setose (Figs 4C/D); frons densely coriaceous (Fig. 4C); hind wing (Fig. 10C) Rs vein almost reaching middle of wing; lower valve with five minute apical teeth..... *C. ichiroi* sp. n. (Fig. 2)

***Cryptalyra belenae* sp. n.**

<http://zoobank.org/11EB3798-3DB0-4238-8254-E5983344B52B>

Figs 1, 4A, B, 5A, B, 6, 9, 12A, B

Material examined. Holotype. Female. BRASIL, Maranhão, Mirador, Parque Est[adual] Mirador, Base dos Cágados, 06°48'29"S, 45°06'34"W, armadilha de Malaise, 27.ix.–02.x.2011, F. Limeira-de-Oliveira, D.W.A. Marques col[etores] (CZMA). Paratype. 1 Female, BRASIL, Maranhão, Carolina, PARNA Chapada das Mesas, Riacho Sucuruí, 240m, 07°07'05.6"S, 47°18'31.6"W, armadilha de Malaise, 10–20.viii.2013, J.A. Rafael, F. Limeira-de-Oliveira, T.T.A. Silva col[etores] (CZMA).

Description. Colour. Wings clear hyaline; tarsi and wings venation light castaneous; mandible, protibia; mesotibia, scape, pedicel, trochanters, femora, and metatibia castaneous; flagellum castaneous with four distal flagellomeres dark castaneous; head, coxae, mesosoma, and metasoma black.

Head (Figs 4A, B, 5A, B). Surface densely foveolate. Compound eye with short and sparse pilosity, with post-ocular sulcus compound by foveae; inner margin divergent posteriorly, in frontal view. Frons with transversal rim on anterior margin. Longitudinal sulcus extends from middle of face to vertex crest. Malar suture indistinct. Gena as long as third part width of eye, with surface uneven and densely foveolate. Occipital carina present; high, longer than half length of gena in profile; strongly scrobiculate throughout and surpassing anterior margin of pronotum laterally; occipital carina at base not curving toward mandible. Palpal formula 5:2.

Mesosoma (Fig. 6). Profemoral depression of pronotum conical; pronotal spiracle minute and semicircular with internal fringe of setae. Mesoscutum medially slightly convex and foveolate and lateral declined area coriaceous; median mesoscutal line (=sul-



Figure 1. *Cryptalyra helenae* sp. n. **A** habitus in lateral view **B** habitus in dorsal view. Scales bar in microns.

cus) deep, broad and scrobiculate; notaulus absent; parapsidal signum absent; mesoscutal humeral sulcus present laterally along entire mesoscutum and scrobiculate; axillae separated medially by its own width, rounded lobe present laterally. Mesoscutellum smooth medially; scutoscuteellar sulcus a row of large foveae; posterior margin of mesoscutellar disc rounded. Metanotum smooth and raised medially, scrobiculate laterally. Propodeum coarsely and irregularly foveolate to areolate; propodeal disc uneven; median longitudinal propodeal sulcus present; postero-lateral corner without tubercle.

Metasoma (Fig. 9A). Cylindrical, tapering posteriorly and longer than mesosoma and head in dorsal view. Abdominal tergum 2 mostly polished. Third and following abdominal tergum coriaceous with a few setae laterally. Ovipositor sheath exceedingly short; ovipositor valves entirely smooth, except four minute teeth on lower valve apically.

Wings (Figs 9B, C). Hind wing with Rs vein almost reaches middle of wing.

Legs (Figs 12A, B). Metacoxa as long as wide, with postero-dorsal surface areolate. Protibia rimmed with stout setae; metatibial setae prone to erect. Basitarsus of all legs conspicuously longer than the other tarsomeres. pro- and mesotibia with one apical spur, metatibia with two apical spurs.

Etymology. The specific epithet is a patronymic honoring Helena Corte Azevedo, daughter of the third author COA.

Remarks. This new species differs from *C. colombia* in the following characters: surface of head densely foveolate; frons with transverse rim on anterior margin, and longitudinal sulcus on its extension; malar suture indistinct; occipital carina finely foveolate dorsally and ventrally; lateral parts of occipital carina curving below and nearly meeting ventrally, not curving towards mandible; mesonotum and axillae shiny but faintly shagreened and deeply pitted with numerous large round foveae, largest mesonotal foveae as broad as head foveae; median mesonotal line formed by continuous fovea, unbounded and deep; propodeum apically areolate-rugose, with longitudinal median sulcus, and posterior margin straight; metacoxa with postero-dorsal region areolate; protibia apically rimmed with stout spines; metatibia with two apical spurs; visible portion of cercus as long as 7th tergite; ovipositor with four minute teeth on lower valve apically; forewing totally hyaline; hind wing with Rs vein reaches middle of wing.

***Cryptalyra ichiroi* sp. n.**

<http://zoobank.org/40AF8EB1-1E77-4244-82C2-11504351A0C2>

Figs 2, 4C, D, 5C, D, 7, 10, 12C, D

Material examined. Holotype. Female. BRASIL, *Maranhão*, Carolina, PARNA Chapada das Mesas, Riacho Cancela, 225m, 07°06'44.2"S, 47°17'56.8"W, armadilha de Malaise, 01–15.vii.2013, J.A. Rafael, F. Limeira-de-Oliveira, T.T.A. Silva col[etores] (CZMA).

Description. Colour. Wings clear hyaline; tarsi light castaneous; mandible, wings venation, protibia and mesotibia castaneous; flagellum castaneous with four distal flag-



Figure 2. *Cryptalyra ichiroi* sp. n. **A** habitus in lateral view **B** habitus in dorsal view. Scales bar in microns.

ellomeres dark castaneous; scape, pedicel, coxae, trochanters, femora, metatibia dark castaneous; head, mesosoma and metasoma black.

Head (Figs 4C, D, 5C, D). Surface uneven and densely coriaceous with few, sparse and unbounded foveae. Compound eye with short and sparse pilosity, without post-ocular sulcus; inner margins parallel in frontal view. Longitudinal sulcus absent on anterior part of head. Malar suture distinct. Gena as long as half width of eye, with surface uneven and densely coriaceous. Occipital carina present; low, shorter than half length of gena in profile; finely foveolate throughout and not surpassing anterior margin of pronotum laterally; occipital carina at base curving toward mandible. Palpal formula 5:2.

Mesosoma (Fig. 7). Pronotum with profemoral depression conical; pronotal spiracle minute and semicircular with internal fringe of setae. Mesoscutum medially slightly convex and coriaceous and lateral declined area coriaceous; median mesoscutal sulcus broad and foveolate; notaulus absent; parapsidal signum absent; mesoscutal humeral sulcus present laterally along entire mesoscutum and slightly scrobiculate; axillae separated medially by its own width, rounded lobe present laterally. Mesoscutellum smooth medially; scutoscuteellar sulcus a row of small foveae; posterior margin of mesoscutellar disc sinuate. Metanotum smooth and raised medially, scrobiculate laterally. Propodeum coarsely and irregularly foveolate to areolate; area of propodeal disc in the same level; median propodeal sulcus absent; postero-lateral corner without tubercle.

Metasoma (Fig. 10A). Cylindrical, tapering posteriorly and longer than mesosoma and head in dorsal view. Abdominal tergum 2 mostly coriaceous with anterior area polished. Third and following abdominal tergum coriaceous with a few setae laterally. Ovipositor sheath exceedingly short; ovipositor valves entirely smooth, except five minute teeth on lower valve apically.

Wings (Figs 10B, C). Hind wing with Rs vein almost reaches middle of wing.

Legs (Figs 12C, D). Metacoxa longer than wide, with longitudinal carina dorsally. Fore- and mesotibia not rimmed with stout setae. Metatibia setae prone to erect. Basitarsus of all legs conspicuously longer than the other tarsomeres. Fore, meso- and metatibia with one apical spur, metatibial spur enclosed.

Etymology. The specific epithet is a patronymic honoring Mateus Ichiro Calhau Kawada, son of the first author RK.

Remarks. This new species differs from *C. plaumanni* in the following characters: body entirely black and wings clear hyaline; eye not setose; malar suture distinct; occipital carina finely foveolate dorsally and ventrally; lateral parts of occipital carina curving below and nearly meeting ventrally, not curving towards mandible; maxillary palp 5-segmented; mesoscutum, axilla and mesoscutellar disc not punctured; pronotum, mesopleuron and metapleuron areolate; propodeum apically areolate-rugose, without distinct tubercles at postero-lateral corner; metacoxa with longitudinal carina extending midway; protibia apically rimmed with stout spines; metasoma cylindrical and coriaceous.



Figure 3. *Cryptalyra limeirai* sp. n. **A** habitus in lateral view **B** habitus in dorsal view. Scales bar in microns.

***Cryptalyra limeirai* sp. n.**

<http://zoobank.org/1590EC1C-199A-4E17-ABAD-EE807C5DEF5D>

Figs 3, 4E, F, 5E, F, 8, 11, 12E, F

Material examined. Holotype. Female. BRASIL, *Maranhão*, Carolina, PARNA Chapada das Mesas, Riacho Cancela, 225m, 07°06'44.2"S, 47°17'56.8"W, armadilha de Malaise, 01–15.vii.2013, J.A. Rafael, F. Limeira-de-Oliveira, T.T.A. Silva col[etores] (CZMA). Paratypes. 1 female, Mirador, Parque Est[adual] Mirador, Base da Geraldina, 419m, 06°37'25"S, 45°52'08"W, armadilha de Malaise, 01–10.x.2013, F. Limeira-de-Oliveira, L.L.M. Santos, T.L. Rocha col[etores] (CZMA); 2 females, 14–18.viii.2012, F. Limeira-de-Oliveira, J.S. Pinto, D.W.A. Marques col[etores] (CZMA); 1 female, BRASIL, *Maranhão*, Carolina, PARNA Chapada das Mesas, Riacho Corrente 288 m 07°04'24.2"S, 47°05'25.2"W, armadilha Malaise, 20–31.viii.2013, J.A. Rafael, F. Limeira-de-Oliveira, T.T.A. Silva col[etores] (CZMA); 1 female, Riacho Cancela, 225m, 07°06'44.2"S, 47°17'56.8"W, 01-15.vii.2013 (CZMA); 1 female, 01-10.x.2013 (CZMA).

Description. Colour. Wings clear hyaline; tarsi and palpi light castaneous; coxae, trochanters, femora, and tibiae castaneous; mandible, scape, pedicel, flagellum, wing venation, and metasoma dark castaneous; head and mesosoma black.

Head (Figs 4E, F, 5E, F). Surface densely foveolate. Compound eye with short and dense pilosity, with post-ocular sulcus foveolate; inner margins divergent dorsally, in frontal view. Frons without transverse rim on anterior margin. Middle of face to vertex crest without longitudinal sulcus. Malar suture indistinct. Gena as long as one third of width of eye, with surface uneven and densely foveolate. Occipital carina present; high, longer than half length of gena in profile; strongly scrobiculate throughout and surpassing anterior margin of pronotum laterally; occipital carina at base not curving toward mandible. Palpal formula 5:3.

Mesosoma (Fig. 8). Profemoral depression of pronotum conical; pronotal spiracle minute and semicircular with internal fringe of setae. Mesoscutum medially slightly convex and foveolate and lateral declined area coriaceous; median mesoscutal line formed by continuous fovea, unbounded and widening posteriorly; notaulus absent; parapsidal signum absent; mesoscutal humeral sulcus present laterally along entire mesoscutum and scrobiculate; axillae separated medially by its own width, rounded lobe present laterally. Mesoscutellum smooth medially; scutoscuteellar sulcus a row of large foveae; posterior margin of mesoscutellar disc rounded. Metanotum smooth and raised medially, scrobiculate laterally. Propodeum coarsely and irregularly foveolate to areolate; area of propodeal disc uneven; median propodeal sulcus absent; postero-lateral corner without tubercle.

Metasoma (Fig. 11A). Cylindrical, tapering posteriorly and longer than mesosoma and head in dorsal view. Abdominal tergum 2 mostly polished. Third and following abdominal tergum coriaceous with a few setae laterally; visible portion of cercus 2.0 × long as 7th tergite; ovipositor sheath exceedingly short; ovipositor valves entirely smooth, except six minute teeth on lower valve apically.

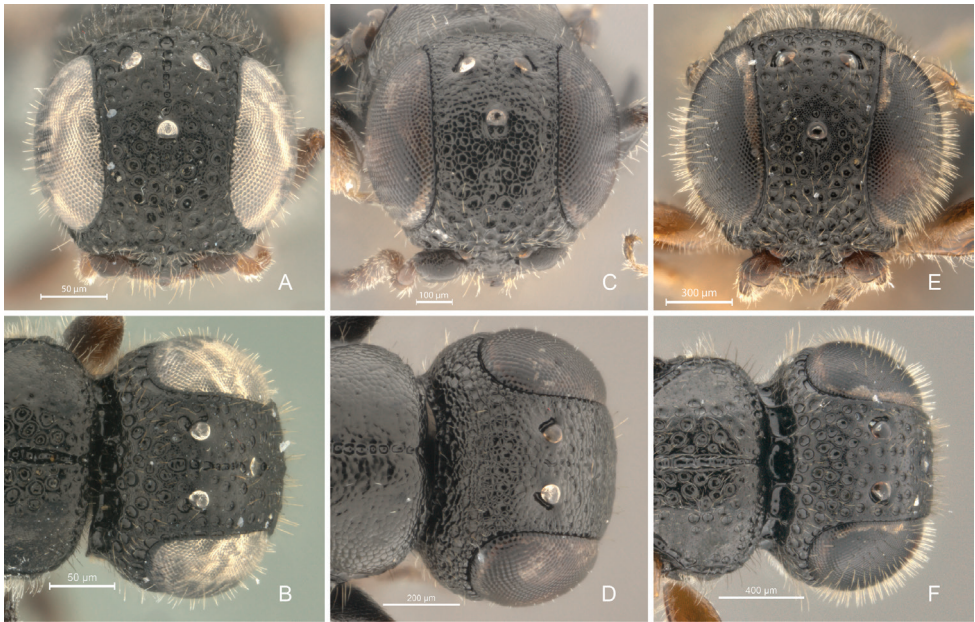


Figure 4. *Cryptalyra helenae* sp. n. **A** head in frontal view **B** head in dorsal view. *Cryptalyra ichiroi* sp. n. **C** head in frontal view **D** head in dorsal view. *Cryptalyra limeirai* sp. n. **E** head in frontal view **F** head in dorsal view. Scales bar in microns.



Figure 5. *Cryptalyra helenae* sp. n. **A** head in ventral view **B** antenna in dorsal view. *Cryptalyra ichiroi* sp. n. **C** head in ventral view **D** antenna in dorsal view. *Cryptalyra limeirai* sp. n. **E** head in ventral view **F** antenna in dorsal view. Scales bar in microns.

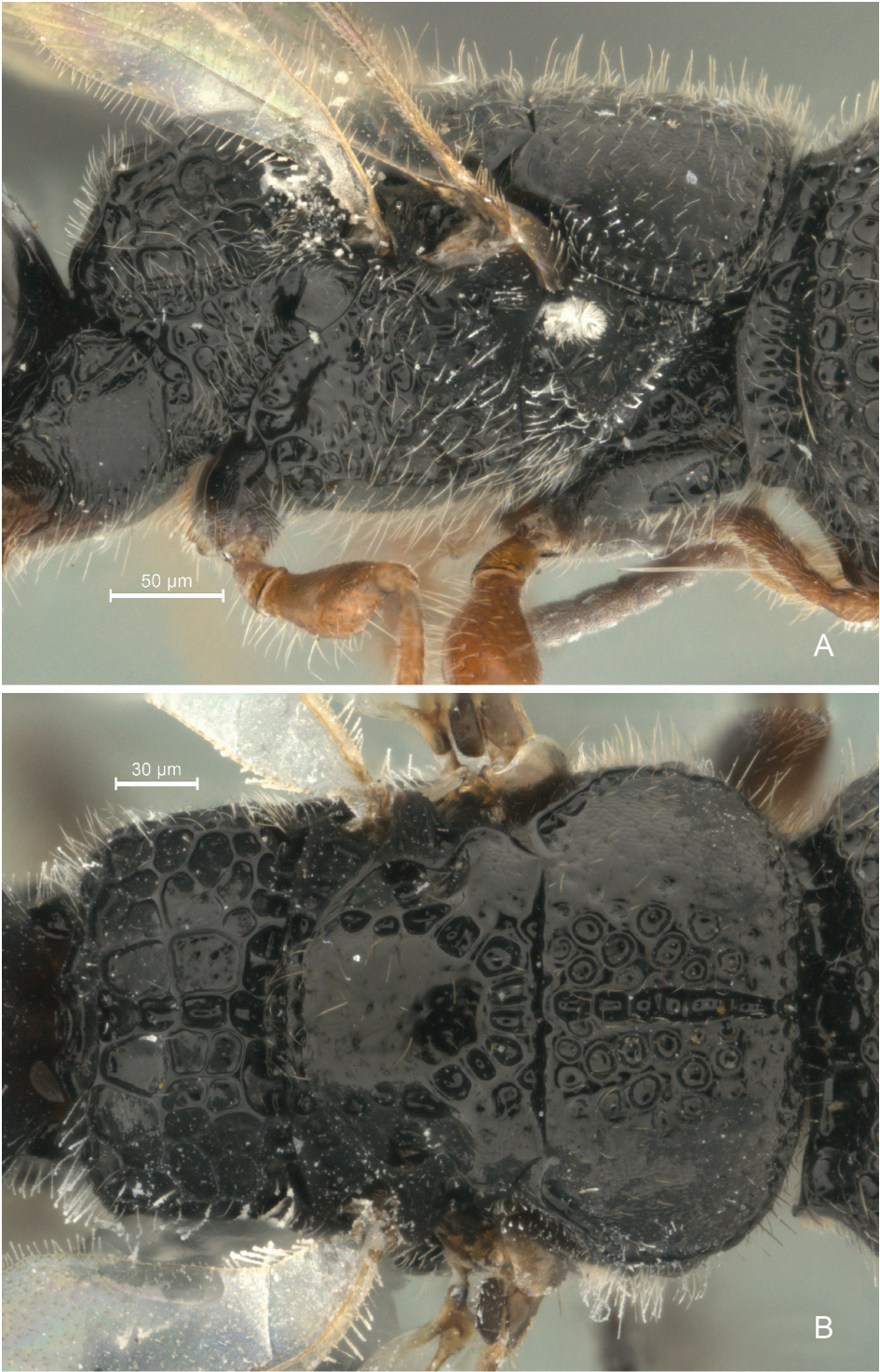


Figure 6. *Cryptalyra helenae* sp. n. **A** mesosoma lateral view **B** mesosoma dorsal view. Scales bar in microns.

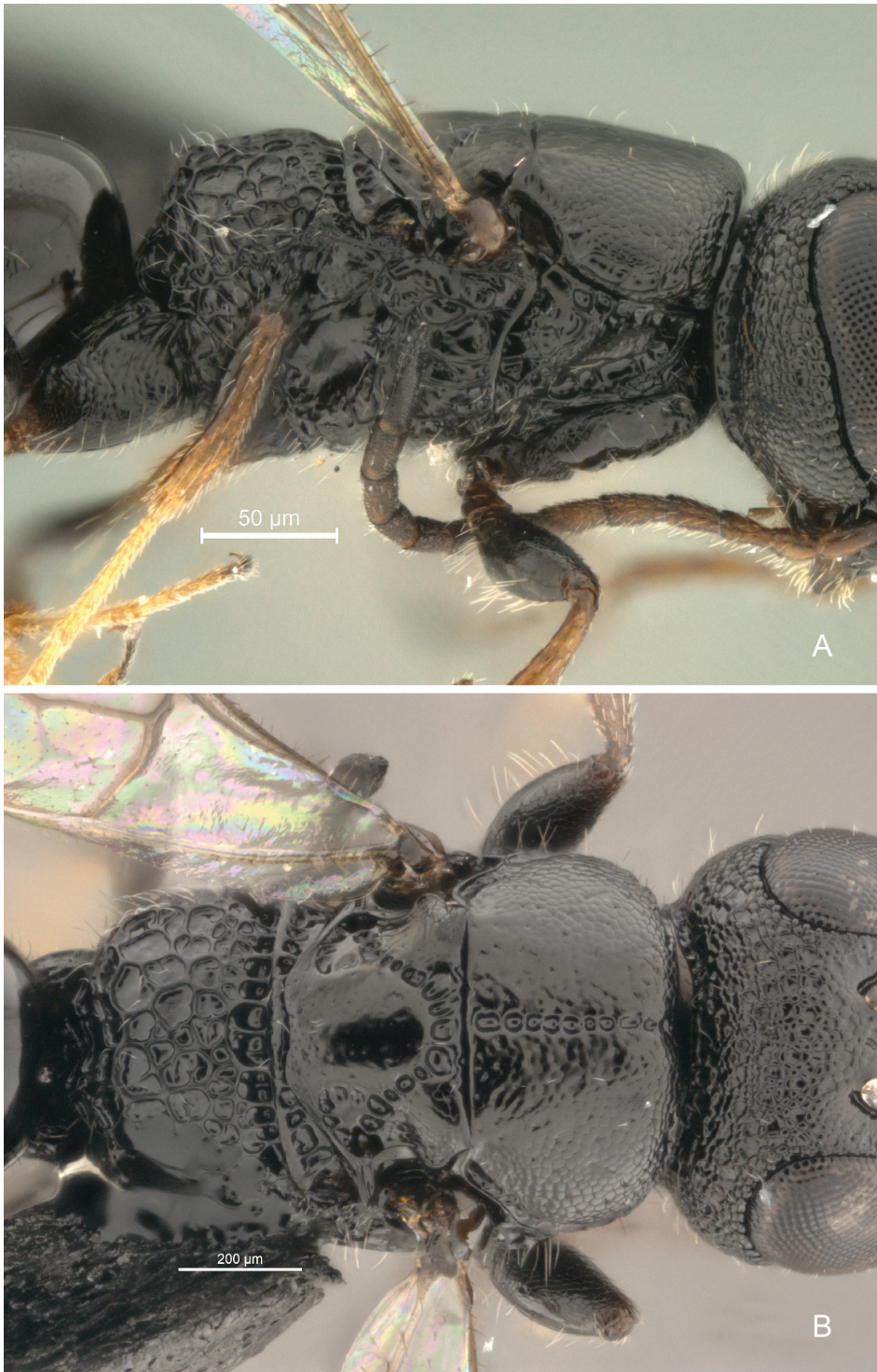


Figure 7. *Cryptalyra ichiroi* sp. n. **A** mesosoma lateral view **B** mesosoma dorsal view. Scales bar in microns.

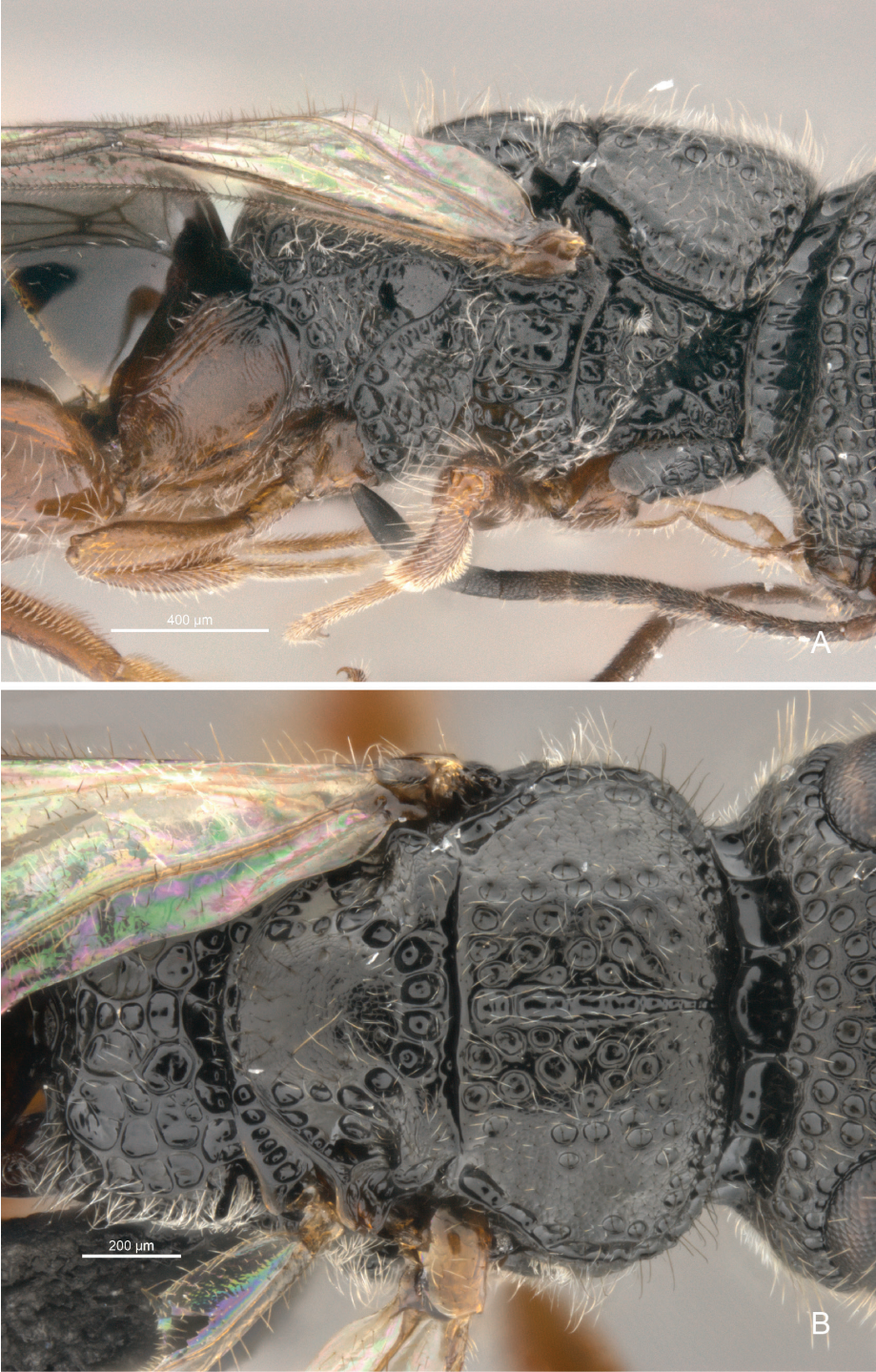


Figure 8. *Cryptalyra limeirai* sp. n. **A** mesosoma lateral view **B** mesosoma dorsal view. Scales bar in microns.

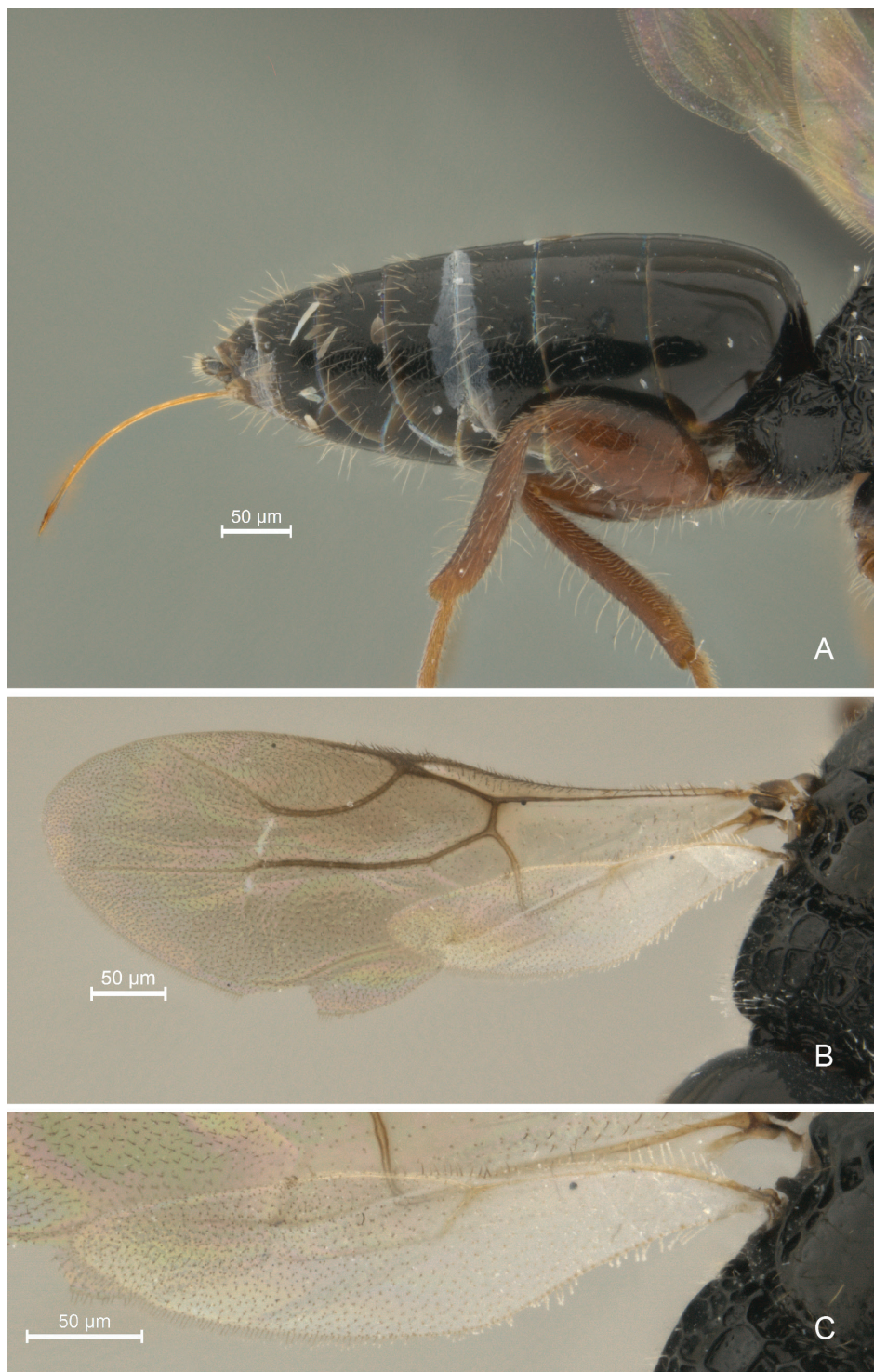


Figure 9. *Cryptalyra helenae* sp. n. **A** metasoma lateral view **B** forewing **C** hind wing. Scales bar in microns.



Figure 10. *Cryptalyra ichiroi* sp. n. **A** metasoma lateral view **B** forewing **C** hind wing. Scales bar in microns.

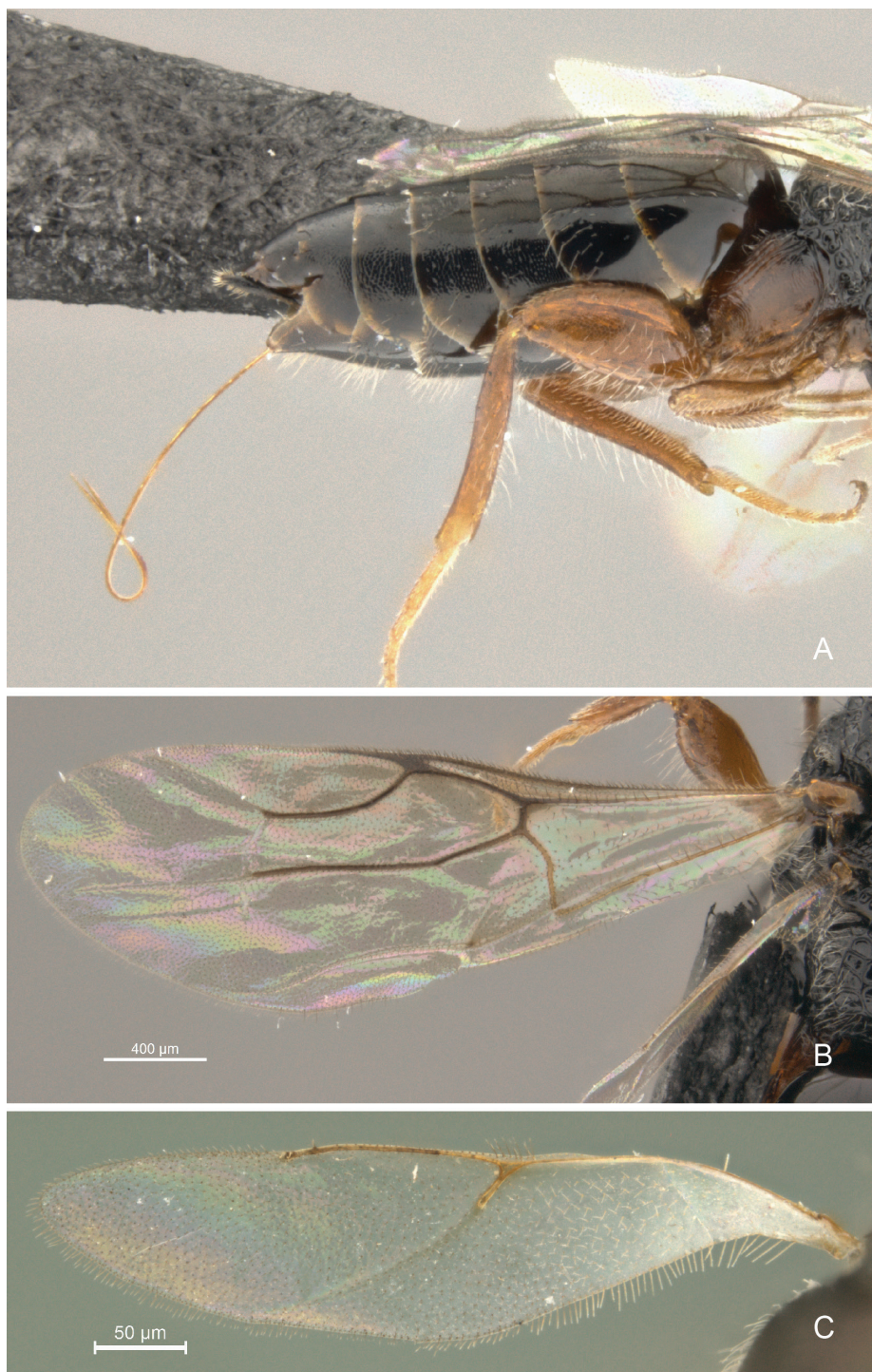


Figure 11. *Cryptalyra limeirai* sp. n. **A** metasoma lateral view **B** forewing **C** hind wing. Scales bar in microns.

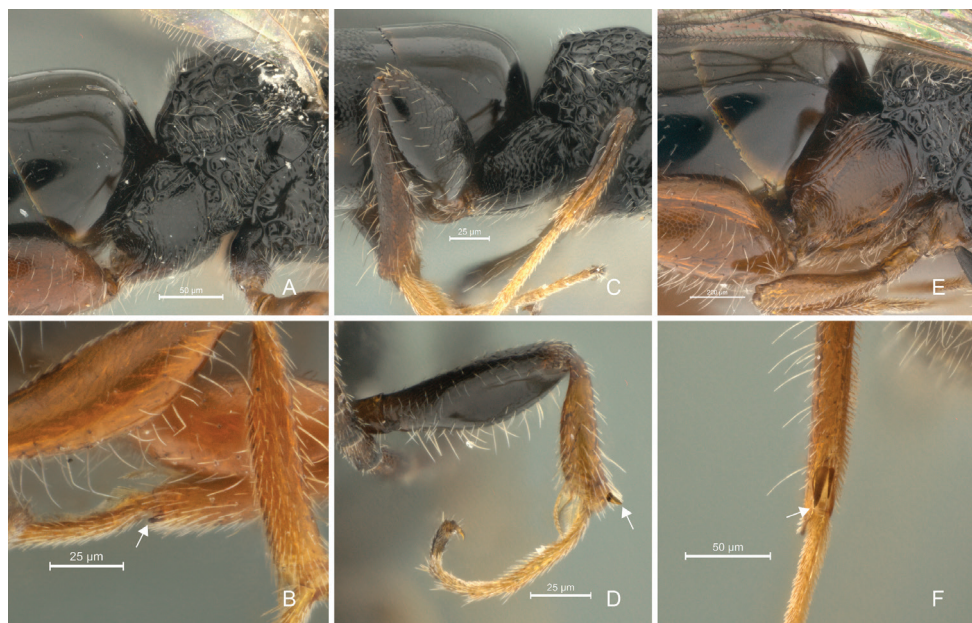


Figure 12. *Cryptalyra helenae* sp. n. **A** metacoxa lateral view **B** stout setae (white arrow) of mesotibia, lateral view. *Cryptalyra ichiroi* sp. n. **C** metacoxa lateral view **D** stout setae (white arrow) of fore leg, lateral view. *Cryptalyra limeirai* sp. n. **E** metacoxa lateral view **F** metatibial spurs (white arrow), posterior view. Scales bar in microns.

Wings (Figs 11B, C). Hind wing with Rs vein almost reaches middle of wing.

Legs (Figs 12E, F). Metacoxa as long as wide, with longitudinal carina dorsally. Pro- and mesotibia rimmed with stout setae; metatibia setae prone to erect. Basitarsus of all legs conspicuously longer than the other tarsomeres. Pro- and mesotibia with one apical spur, metatibia with two apical spurs.

Etymology. The specific epithet is a patronymic honoring Francisco Limeira-de-Oliveira, Diptera researcher and curator of CZMA.

Remarks. This new species differs from *C. colombia* in the following characters: lateral parts of occipital carina curving below and nearly meeting ventrally, curving towards mandible; median mesoscutal line formed by continuous fovea, unbounded and widening posteriorly; propodeum coarsely and irregularly areolate; pro- and mesotibia apically rimmed with stout spines; metatibia with two apical spurs; visible portion of cercus 2.0 × long as 7th tergite; ovipositor six minute teeth on lower valve apically; forewing totally hyaline; hind wing with Rs vein reaches middle of wing.

Discussion

Here we increased the number of described species of *Cryptalyra* from three to six. Several new character conditions broadened the genus concept: the metatibia of *C.*

helenae has two spurs in rather than one as in other species (only one spur supposed to be present in *Cryptalyra* spp., see key in Vilhelmsen et al. 2010); the head of *C. helenae* has a longitudinal sulcus which runs medially from the middle of face to vertex crest (Figs 4A, B); the sulcus is like a line of foveae which is stronger on the vertex than on the face, this sulcus is not found in the other species. The mesonotum of *C. limeirai* is continuous (Fig. 8B) rather than formed by foveae as in the other species such as *C. ichiroi* (Fig. 7B). The propodeal disc of *C. helenae* has a median longitudinal sulcus (Fig. 6B), this sulcus is not present in the other species, however *C. depressa* presents a longitudinal wide depression characterizing this species, not being the same as found in *C. helenae*. The dorsum of metacoxa is usually carinate (Figs 12C, E), but it is scrobiculate in *C. helenae* (Fig. 12A). The ovipositor has usually three teeth on the dorsal side of the valve, however the *C. helenae* ovipositor has four teeth and *C. limeirai* and *C. ichiroi* have six.

Cryptalyra was recorded from Caquetá in southern Colombia (Shaw 2003), Santa Catarina in southern Brazil (Shaw 1987) and Pará in Northern Brazil (Azevedo and Tavares 2006). These three new species here described are from Maranhão, Brazil, which is relatively close to the latter site, about 460 km in a straight line. However, the three previous nominal species were collected in rain forest and these new ones were collected in savannah, a place much drier than rain forest, although there is a regular wet season.

This genus was known only from three specimens. Now we have ten specimens, representing three times more specimens. They were collected under the scope of two projects, both coordinated by Francisco Limeira-de-Oliveira. They are “Riqueza, diversidade e composição de insetos do Parque Estadual do Mirador, Maranhão, Brasil” and “Diversidade de Díptera dos Parques Nacionais Chapada das Mesas e Serra das Confusões”. The total sampling effort was 2,700 days of Malaise trap at Parque Estadual do Mirador and 4,320 days of Malaise trap at Parque Nacional Chapada das Mesas. The traps were continuously set up in the field. Interestingly, according to Lars Vilhelmsen (pers. comm.) there are two recently collected specimens of *Cryptalyra* from French Guiana in the collection of the Natural History Museum of Copenhagen. Using the key presented herein he identified that material as apparently belonging to *C. depressa* and *C. ichiroi*, respectively. These specimens were also collected as a result of a continuous collecting effort, by the Société entomologique Antilles-Guyane (SEAG). As we can see, this strategy has shown to be very effective for Megalyridae. Although we have only one year of collecting in the field, continuous sampling can be useful to reveal potential seasonality. All ten specimens of *Cryptalyra* were collected from July to October.

The state of Maranhão in Brazil is here demonstrated to be a diversity spot for *Cryptalyra*. It is mostly covered by savannah, but its northwestern area is covered by Amazon rain forest, and the eastern border is covered by Caatinga vegetation. The confluence of these three main ecosystems makes Maranhão a place with huge potential for new discovery of biodiversity, especially Hymenoptera parasitoids, a group understudied in the Neotropics.

Acknowledgment

We are thank to Francisco Limeira-de-Oliveira for his kind reception at Coleção Zoológica do Maranhão in Caxias; to Scott Shaw for providing papers and comments; to CNPq grant #151153/2013-2, FAPES grant #2012/20 and CNPq grant #301669/2010-4 for providing scholarship to first, second and third authors respectively. The two trips to Caxias were supported by CNPq/FAPES grant #52263010/2011 (Pronex) and CNPq grant #562224/2010-6 (Programa Taxonomia).

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Hisonotus acuen, a new and phenotypically variable cascudinho (Siluriformes, Loricariidae, Hypoptopomatinae) from the upper rio Xingu basin, Brazil

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Abstract

A new species of *Hisonotus* is described from the headwaters of the rio Xingu. The new species is distinguished from its congeners by having a functional V-shaped spinelet, odontodes not forming longitudinal aligned rows on the head and trunk, lower counts of the lateral and median series of abdominal plates, presence of a single rostral plate at the tip of the snout, absence of the unpaired platelets at typical adipose fin position, yellowish-tipped teeth, absence of conspicuous dark saddles and stripe on the body and higher number of teeth on the premaxillary and dentary. The new species, *Hisonotus acuen*, is restricted to headwaters of the rio Xingu basin, and is the first species of the genus *Hisonotus* described from the rio Xingu basin. *Hisonotus acuen* is highly variable in aspects of external body proportions, including body depth, snout length, and abdomen length. This variation is partly distributed within and among populations, and is not strongly correlated with body size. PCA of 83 adult specimens from six allopatric populations indicates the presence of continuous variation. Therefore, the available morphological data suggest that the individuals inhabiting the six localities of rio Xingu represent different populations of a single species. Low intraspecific variation in mitochondrial Cytochrome oxidase subunit I (COI) provides corroborative evidence.

Keywords

Biodiversity, Cascudinhos, Fresh-water, Neotropical fish, Taxonomy

Introduction

The subfamily Hypoptopomatinae is a monophyletic group of Loricariids (Schaefer, 2003) composed of 19 genera and 139 species (Eschmeyer and Fong 2014). Within this subfamily, *Hisonotus* Eigenmann & Eigenmann, 1889 comprises 33 valid species (Eschmeyer 2014). The genus *Hisonotus* was resurrected from the synonymy of *Otocinclus* by Schaefer (1998) based on the reduced or absent snout plates anterior to the nostril, rostrum with enlarged odontodes, and thickened plates forming the lateral rostral margin. However, the phylogenetic relationships in this genus are not well resolved (Britski and Garavello 2007) and, according to molecular (Chiachio et al. 2008; Cramer et al. 2011) and morphological (Martins et al. 2014) data, *Hisonotus* is a polyphyletic genus.

Although there is no definition of *Hisonotus* that supports its monophyly, many authors have considered this genus as valid. In the past decade, 18 species of *Hisonotus* have been described (Britski and Garavello 2007; Carvalho et al. 2008; Carvalho and Reis 2009; Carvalho and Reis 2011; Martins and Langeani 2012; Carvalho and Datovo 2012; Roxo et al. 2013; Roxo et al. 2014). Recently, during a collecting trip in tributaries of the rio Xingu basin, we found fish specimens that have the generally accepted characteristics of *Hisonotus* listed above but do not match any known species. Herein we describe the rio Xingu specimens as a new species.

Material and methods

All measurements and counts were taken on the left side of specimens. Measurements were taken point to point to the nearest 0.1 mm with a digital caliper. Body plate and osteology nomenclature followed Schaefer (1997) and measurements followed Carvalho and Reis (2009), as shown in Table 1. Abbreviations used in the text followed Carvalho and Reis (2009). Morphometrics are given as percentages of standard length (SL), except for subunits of the head region, which are expressed as percentages of head length (HL). Specimens were cleared and stained (c&s) according to the method of Taylor and Van Dyke (1985). Vertebral counts also include the five vertebrae that comprise the Weberian apparatus. Dorsal-fin ray counts include the spinelet as the first unbranched ray. All examined specimens were collected according to the Brazilian laws, and are deposited under permanent scientific collection licenses. After collection, specimens were euthanized using 1% benzocaine in water, fixed in 10% formaldehyde for morphological studies and preserved in 70% alcohol. For molecular studies specimens were fixed directly in 95% alcohol. Sequencing and molecular analysis followed Roxo et al. (2012). Institutional acronyms follow Fricke and Eschmeyer (2014). All samples are deposited at the LBP, Laboratório de Biologia e Genética de Peixes, Universidade Estadual Paulista, Botucatu; MZUSP, Museu de Zoologia, Universidade de São Paulo, São Paulo; NUP, Coleção Ictiológica do Nupelia, Universidade Estadual de Maringá, Maringá. Zoological nomenclature

follows the International Code of Zoological Nomenclature (4th Ed.). The GenBank accession numbers for Cytochrome oxidase subunit I (COI) sequences are: *H. acuen* – KM365043, KM365044, KM365045, KM365046, KM365047, KM365048, KM365049, KM365050, KM104473; *H. chromodontus* – KM104474, KM104475, JN998567, JN998566, JN998567, JN998565, JN998564, KM365054; *H. insperatus* – KM104485, GU701888, GU701749, GU701748, GU701747, GU701746, KM365055, KM365056, KM365057, KM365058, KM365059, KM365060, KM365061; *H. notatus* – JN998579, JN998581, JN998580; *H. oliveirai* – KM104486, KM365062, KM365063; *H. paresi* – KM104490, KM365042; *H. piracanjuba* – KM104487, KM104488, KM365051, KM365052, KM365053.

Principal component analysis (PCA)

Principal component analysis (PCA) was used to check overall variation among samples, including differences in morphometrics among species. PCA is a statistical procedure that uses orthogonal transformation to convert a set of observations of possibly correlated variables into a set of values of linearly uncorrelated variables called principal components (Jolliffe 2002). The analyses were made using all measurements listed above. Juvenile specimens below 18.0 mm SL were excluded from the analyses. PCA on covariances of base 10 logarithmically transformed measurements to reduce the influence of size were obtained using Past version 1.28 (Hammer et al. 2004). The PCA Loadings are presented in Table 2.

Results

Hisonotus acuen sp. n.

<http://zoobank.org/12454B4D-E9CA-4A89-A307-B77A703308C9>

Figs 1, 5, 6; Table 1

Holotype. MZUSP 115350, female, 25.9 mm SL, Brazil, Mato Grosso State, municipality of Querência, affluent of rio Toguro, rio Xingu basin, 13°00'26"S, 52°11'27"W, 01 Aug 2012, coll. C. Oliveira, M. Taylor, G.J.C. Silva & J.M. Henriques.

Paratypes. All from Brazil, Mato Grosso State, rio Xingu basin. LBP 15755, 16, 19.5–26.0 mm SL, municipality of Ribeirão Cascalheira, affluent of rio Suiá-Missu, 12°55'36"S, 51°53'27"W, 30 July 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16274, 27, 20.2–29.1 mm SL, 2 c&s 23.6–24.2 mm SL, municipality of Gaúcha do Norte, affluent of rio Culuene, 13°27'26"S, 53°09'36"W, 03 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16275, 29, 16.7–25.2 mm SL, 2 c&s 19.3–20.8 mm SL, municipality of Querência, affluent of rio Feio, 12°33'20"S, 52°16'16"W, 31 Sep 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16276, 9, 20.7–27.9 mm SL, 2 c&s 21.2–21.4 mm SL, munici-

Table 1. Morphometrics and meristic data for all analyzed specimens of *Hisonotus acuen* and for specimens by sub basins. SD = Standard deviation.

	<i>Hisonotus acuen</i> , n = 83					rio Toguero, n = 7					rio Culcuene, n = 29					rio Suía Missu, n = 15				
	Holotype	Low	High	Mean	SD	Low	High	Mean	SD	Low	High	Mean	SD	Low	High	Mean	SD			
SL	25.9	18.2	29.0	23.3	2.6	20.5	25.9	22.9	2.23	21.7	29.0	25.6	1.7	20.5	27.1	23.3	2.0			
Percents of SL																				
Head length	39.4	35.1	44.1	39.4	1.8	38.4	44.1	40.7	1.7	8.81	10.8	9.7	0.4	8.3	10.5	9.3	0.6			
Predorsal length	50.1	41.9	54.3	50.0	1.9	48.6	52.8	51.1	1.5	41.9	54.3	48.8	2.0	49.1	52.7	51.1	1.1			
Dorsal-fin spine length	20.6	19.4	25.8	21.9	1.4	19.5	22.8	20.7	1.06	4.9	6.3	5.6	0.3	4.0	6.2	5.0	0.6			
Anal-fin unbranched ray length	15.3	13.7	19.9	16.9	1.3	14.9	16.6	15.7	0.69	3.8	5.0	4.4	0.2	2.8	4.2	3.6	0.3			
Pectoral-fin spine length	24.0	15.9	28.8	25.0	1.9	21.6	26.6	23.9	1.68	22.7	28.8	25.6	1.4	4.9	6.6	5.8	0.5			
Pelvic-fin unbranched ray length	16.1	13.1	25.0	16.9	2.1	13.9	17.6	15.8	1.17	15.2	23.2	17.6	2.1	2.8	4.1	3.6	0.3			
Cleithral width	22.1	13.2	27.5	23.9	1.9	22.1	24.6	23.6	1.05	5.2	7.0	6.3	0.5	4.6	6.2	5.4	0.5			
Thoracic length	12.8	10.5	23.1	13.7	2.0	11.2	16.5	13.4	2.07	2.8	4.5	3.7	0.4	2.2	5.8	3.2	0.9			
Abdominal length	22.0	10.2	24.4	20.8	2.5	18.3	22.0	20.6	1.16	3.6	6.6	5.6	0.6	2.1	6.0	4.5	1.1			
Caudal-peduncle length	27.7	25.5	33.0	28.6	1.5	26.1	30.0	27.7	1.18	6.3	8.6	7.4	0.6	5.2	7.8	6.5	0.7			
Caudal-peduncle depth	9.5	8.6	11.1	9.6	0.4	8.7	10.3	9.5	0.54	8.9	11.1	9.9	0.4	9.0	10.3	9.5	0.4			
Percents of HL																				
Snout length	56.0	34.2	57.2	53.5	2.6	41.2	56.0	51.8	4.95	51.5	56.7	53.6	1.52	34.2	56.4	53.0	5.4			
Orbital diameter	13.4	11.2	16.2	13.1	0.9	11.6	13.8	12.7	0.83	11.7	15.0	13.4	0.8	1.0	1.3	1.1	0.1			
Interorbital width	36.2	15.6	41.8	36.3	3.4	33.1	53.0	37.1	1.05	3.1	4.1	3.7	0.2	2.7	3.9	3.3	0.3			
Head depth	51.7	35.1	53.1	44.2	4.6	40.0	56.5	45.5	6.1	3.9	5.3	4.6	0.3	3.2	4.8	4.0	0.5			
Suborbital depth	19.2	13.3	22.4	17.4	2.2	12.6	19.2	16.4	2.3	16.0	21.0	19.0	1.3	1.1	1.8	1.5	0.2			
Mandibular ramus	11.5	6.9	12.9	10.2	1.4	9.7	17.9	12.5	3.6	8.3	12.9	10.9	1.3	8.5	9.6	9.1	0.4			
Meristics																				
Left premaxillary teeth	23	14	27	14	4.1	14	20	-	3.87	22	24	23.3	0.8	14	21	17.2	2.5			
Left dentary teeth	18	12	23	21	3.5	13	18	-	2.38	12	23	21	3.5	13	17	15.0	1.5			
Left lateral scutes	23	22	24	23	0.7	23	24	23	0.82	20	23	21.3	1.0	24	24	24	-			

Table 1. Continued.

	córrego Xavante, n = 13				rio Coronel Vanick, n = 4				rio Feio, n = 15			
	Low	High	Mean	SD	Low	High	Mean	SD	Low	High	Mean	SD
SL	19.4	25.6	22.1	2.0	19.5	24.8	21.5	2.4	18.2	24.6	20.8	1.9
Percents of SL												
Head length	37.2	41.7	39.1	1.2	38.2	41.2	39.9	1.3	39.2	42.7	41.2	1.1
Predorsal length	47.5	51.5	49.2	1.1	49.1	52.9	51.1	1.6	49.0	52.9	51.3	1.3
Dorsal-fin spine length	20.2	24.7	22.2	1.1	19.4	21.6	20.5	0.7	20.0	24.4	22.3	1.1
Anal-fin unbranched ray length	16.4	19.9	17.6	0.8	13.9	16.7	15.7	1.2	15.5	19.7	17.7	1.0
Pectoral-fin spine length	15.9	27.7	24.5	3.6	23.2	25.6	24.8	1.0	23.3	26.8	24.9	1.0
Pelvic-fin unbranched ray length	15.5	20.7	17.8	1.7	15.6	16.3	15.9	0.3	13.1	25.0	16.8	2.8
Cleithral width	23.9	27.5	25.5	0.8	13.2	25.4	22.0	5.8	20.8	22.9	21.8	0.6
Thoracic length	10.5	16.6	12.8	1.5	11.6	13.5	12.7	0.8	11.2	19.0	12.9	2.0
Abdominal length	12.1	22.5	20.3	2.5	20.3	22.1	21.2	0.7	18.4	23.9	20.3	1.3
Caudal-peduncle length	27.1	32.1	29.2	1.3	26.2	27.6	26.7	0.6	26.8	30.9	28.8	1.4
Caudal-peduncle depth	9.1	10.3	9.7	0.3	9.1	9.7	9.4	0.2	8.6	9.9	9.2	0.3
Percents of HL												
Snout length	50.3	54.6	52.9	1.1	51.3	53.7	52.7	1.0	52.7	57.2	54.9	1.24
Orbital diameter	12.2	16.2	13.7	1.1	12.5	14.2	13.4	0.7	11.2	14.6	13.1	0.9
Interorbital width	35.8	41.8	38.5	1.6	33.8	36.8	35.1	1.2	15.6	37.6	32.9	5.1
Head depth	40.6	51.4	46.5	3.0	40.2	43.8	41.6	1.5	35.1	39.9	37.5	1.5
Suborbital depth	14.7	22.4	19.1	2.0	14.4	18.4	16.1	1.9	13.3	17.5	14.9	1.2
Mandibular ramus	7.3	10.1	8.8	1.0	7.2	8.9	8.1	0.4	6.9	11.5	9.3	1.2
Meristics												
Left premaxillary teeth	20	27	-	2.94	17	25	-	4.0	14	15	14	0.5
Left dentary teeth	16	22	-	2.75	14	17	-	1.5	12	13	12	0.5
Left lateral scutes	23	24	23	0.5	23	24	23	0.5	22	24	22	1.5

Table 2. Variable loadings in the first and second axes of size-free Principal Component Analysis (Axis 1 and Axis 2) of combined samples of *H. acuen*.

	Axis 1	Axis 2
Predorsal length	0.1871	-0.0346
Preanal length	0.2139	-0.0923
Head length	0.1631	-0.0090
Cleithral width	0.2370	0.1401
Dorsal-fin spine length	0.2015	0.0902
Base of dorsal-fin length	0.2838	-0.2170
Thorax length	0.2649	-0.1924
Pectoral-fin spine length	0.2234	0.0939
Abdomen length	0.2688	-0.5397
Pelvic-fin spine length	0.2155	0.4822
Anal-fin spine length	0.1747	0.2453
Lower cd spine	0.2088	0.0789
Caudal peduncle depth	0.1887	0.0911
Caudal peduncle length	0.2168	0.3770
Anal width	0.2543	-0.1638
Body depth	0.2608	-0.2413
Head depth	0.2506	0.0613
Snout length	0.1376	-0.1066
Interorbital width	0.2147	0.1441
Orbital diameter	0.1189	0.0778
Suborbital depth	0.2368	0.0426

pality of Ribeirão Cascalheira, affluent of rio Suiá-Missu, 12°53'04"S, 52°02'00"W, 30 Sep 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16277, 10, 18.9–23.3 mm SL, municipality of Querência, affluent of rio Feio, 12°31'55"S, 52°20'29"W, 31 Sep 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16278, 12, 18.8–25.1 mm SL, 2 c&s 26.8–27.1 mm SL, municipality of Primavera do Leste, córrego Xavante, 14°38'24"S, 53°55'38"W, 05 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16279, 10, 20.8–26.7 mm SL, municipality of Gaúcha do Norte, affluent of rio Culuene, 13°26'32"S, 53°08'45"W, 03 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16280, 11, 17.4–24.9 mm SL, municipality of Canarana, affluent of rio Culuene, 13°25'30"S, 52°16'47"W, 01 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16281, 4, 17.5–24.6 mm SL, same collection information as holotype. LBP 16282, 5, 17.5–23.9 mm SL, municipality of Canarana, rio Coronel Vanick, 13°31'34"S, 52°43'52"W, 02 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16283, 2, 21.3–24.2 mm SL, municipality of Canarana, affluent of rio Toguro, 13°16'52"S, 52°14'42"W, 01 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. LBP 16284, 3, 20.2–24.3 mm SL, collected with holotype. LBP 18845, 1, 23.7 mm SL, municipality of Gaúcha do Norte, affluent of rio Culuene, 13°30'57"S, 53°06'39"W, 03 Aug



Figure 1. *Hisonotus acuen*, MZUSP 115350, female, 25.9 mm SL, holotype, from Mato Grosso State, municipality of Querência, affluent of rio Toguro, rio Xingu basin, 13°00'26"S, 52°11'27"W.

2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM. NUP 16444, 5, 22.2–27.1 mm SL, municipality of Gaúcha do Norte, affluent of rio Culuene, 13°27'26"S, 53°09'36"W, 03 Aug 2012, coll. Oliveira C, Taylor M, Silva GJC, Henriques JM.

Diagnosis. *Hisonotus acuen* differs from all congeners except *H. bockmanni*, *H. chromodontus*, *H. insperatus*, *H. luteofrenatus*, *H. oliveirai* and *H. parsi* by having a functional V-shaped spinelet, Fig. 2a–f (*vs.* non-functional spinelet, a square ossification, or spinelet absent, Fig. 2g–h). It differs from *H. insperatus*, *H. parsi*, *H. luteofrenatus*, and

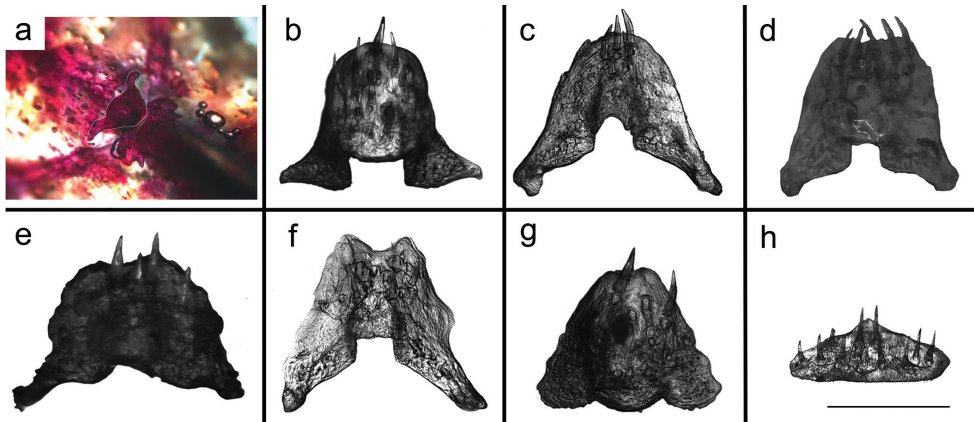


Figure 2. Spinelet variation among *Hisonotus* species. Figure a–f showing the extension of the bone forming a lock mechanism. In figure g–h, the bones lack the lock mechanism: **a** *H. acuen*, LBP 16276, 21.4 mm SL **b** *H. acuen*, LBP 16274, 21.1 mm SL **c** *H. oliveirai* LBP 13332, 23.7 mm SL **d** *H. chromodontus*, LBP 7964, 28.3 mm SL **e** *H. piracanjuba*, LBP 17256, 27.1 mm SL **f** *H. paresis*, NUP 10928, 23.6 mm SL **g** *H. cf. notatus*, LBP 3472, 25.8 mm SL **h** *H. depressicauda*, LBP 17474, 28.1 mm SL. Scale bar = 0.5 mm.

H. oliveirai by having odontodes not forming longitudinally aligned rows on head and trunk (*vs.* odontodes forming longitudinally aligned rows on head and trunk). *Hisonotus acuen* differs from *H. insperatus* and *H. luteofrenatus* by the lower counts of the lateral series of abdominal plates (4–5 *vs.* 6–8 and 7–8, respectively) and from *H. insperatus* by the lower counts of the lateral median plates (22–24 *vs.* 25–26). The new species can be distinguished from *H. luteofrenatus*, *H. oliveirai* and *H. paresi* by the presence of a single rostral plate at tip of snout (*vs.* presence of a pair of rostral plates at tip of snout); from *H. bockmanni* by the absence of unpaired platelets at typical adipose fin position (*vs.* presence of the unpaired platelets); from *H. chromodontus* by having yellowish-tipped teeth, Fig. 3a (*vs.* reddish-brown teeth, Fig. 3b), by having the caudal-fin color pattern mostly hyaline, except for dark blotch on origin of rays, and dark brown chromatophores largely concentrated on rays near lower caudal spine, Fig. 3c (*vs.* caudal-fin mostly dark brown with chromatophores largely concentrated on rays and membranes, and with two hyaline spots on middle of the fin, Fig. 3d); from *H. paresi* by the absence of conspicuous dark dorsal saddle and longitudinal stripe on the body (*vs.* inconspicuous dark saddles and stripe of the body) and from *H. insperatus* by the higher number of premaxillary (14–27 *vs.* 6–12) and dentary teeth (12–23 *vs.* 5–11).

Description. Morphometric and meristic data presented in Table 1. Maximum body length 29.0 mm SL. Dorsal profile of head in lateral view convex to straight from upper part of rostrum to posterior margin of nares, slightly curved from eyes to posterior margin of parieto supraoccipital, almost straight to dorsal-fin origin. Dorsal profile of trunk almost straight, descending from base of dorsal-fin origin to caudal peduncle. Ventral profile slightly concave from snout tip to anal-fin origin, slightly convex to caudal peduncle. Greatest body depth at dorsal-fin origin (13.5–22.8% SL). Greatest body width at cleithral region, gradually decreasing towards snout and caudal

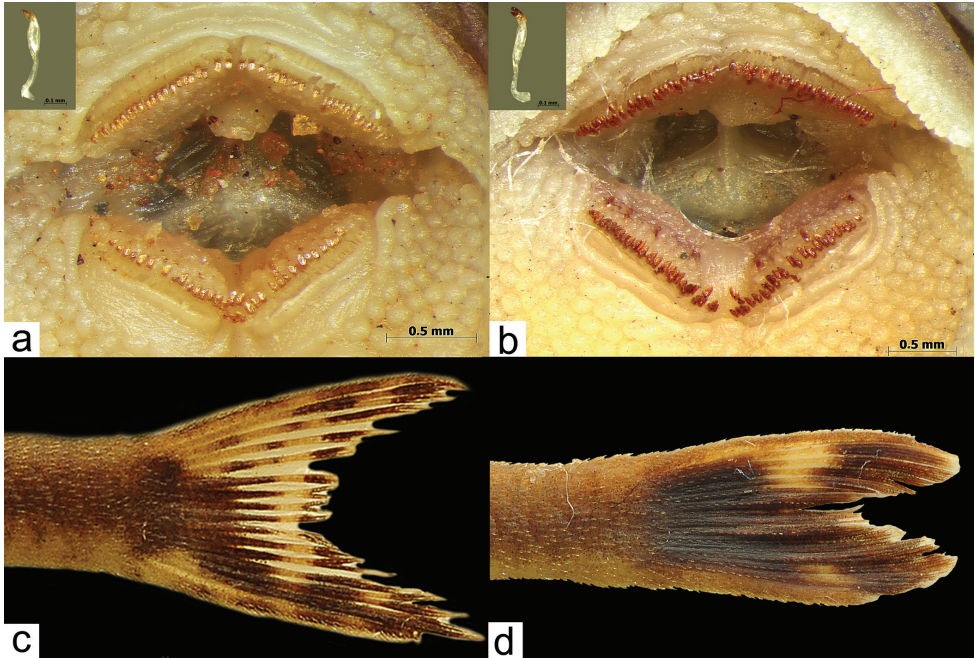


Figure 3. **a** *H. acuen*, holotype, MZUSP 115350, 25.9 mm SL, showing the yellowish-tipped teeth **b** *H. chromodontus*, NUP 10924, 29.7 mm SL, showing the reddish-brown teeth **c** *H. acuen*, holotype, MZUSP 115350, 25.9 mm SL, showing the caudal-fin color pattern mostly hyaline, except for dark blotch on origin of rays, and dark brown chromatophores largely concentrated on rays near lower caudal spine **d** *H. chromodontus*, LBP7964, 27.3 mm SL, showing caudal-fin dark brown with chromatophores largely concentrated on rays and membranes, and with two hyaline spots on middle of the fin.

fin. Cross-section of caudal peduncle almost ellipsoid; rounded laterally and almost flat dorsally and ventrally.

Head rounded in dorsal view. Snout slightly pointed, its tip rounded, elongated (34.2–57.2% HL) and depressed in front of each nostril on dorsal surface. Dorsal and ventral series of odontodes completely covering anterior margin of snout; odontodes of snout similar in size to remaining ones found on head. Snout tip lacking band devoid of odontodes. Odontodes on head and trunk well defined and not forming longitudinal rows. Usually no tufts or crests of odontodes on head, in some juvenile specimens, a tiny tuft of odontodes at posterior tip of supraoccipital. Eyes small (11.2–16.2% HL), dorsolaterally positioned. Lips roundish and papillose; papillae uniformly distributed on base of dentary and premaxilla and slightly decreasing in size distally. Lower lip larger than upper lip; its border fringed. Maxillary barbel present. Teeth slender and bicuspid; mesial cusp larger than lateral cusp. Premaxillary teeth 14–27. Dentary teeth 12–23.

Dorsal-fin ii,7; dorsal-fin spinelet short and V-shaped; dorsal-fin lock functional; its origin slightly posterior to pelvic-fin origin. Tip of adpressed dorsal-fin rays slightly surpassing end of anal-fin base. Pectoral-fin i,6; tip of longest pectoral-fin ray almost reaching half of pelvic-fin length, when depressed. Pectoral axillary slit present be-

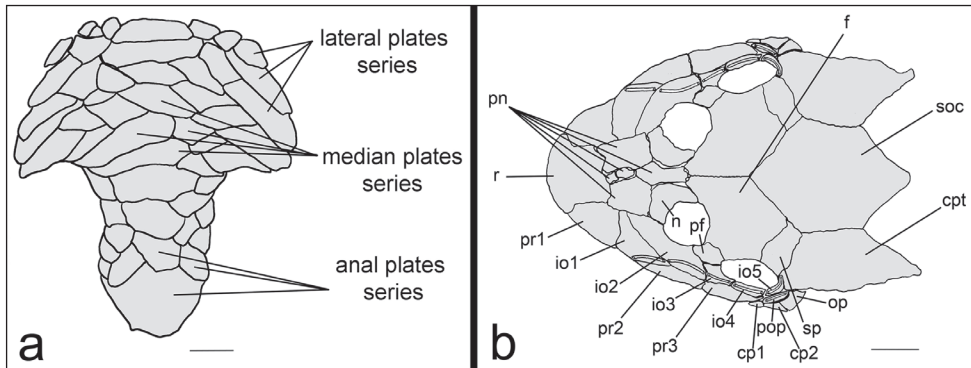


Figure 4. *Hisonotus acuen*, paratype, LBP 16278, 27.1 mm SL **a** Ventral view of abdominal plates **b** Cranial bones and dermal plates of the head in dorsal view. Scale bars: 1 mm.

tween pectoral-fin insertion and lateral process of cleithrum. Pectoral spine supporting odontodes anteroventrally. Pelvic-fin i,5; its tip not exceeding anal-fin origin when depressed in both sex. Pelvic-fin unbranched ray with dermal flap along its dorsal surface in males. Anal fin i,5; its tip reaching 7th and 8th plate from its origin. Caudal-fin i,14,i; distal margin forked. Adipose-fin absent. Total vertebrae 27.

Body covered with bony plates except on ventral part of head, around pectoral and pelvic-fin origin and on dorsal-fin base. Cleithrum and coracoid totally exposed. Arrector fossae partially enclosed by ventral lamina of coracoids. Abdomen entirely covered by plates in adults (about 23.0 mm SL); lateral plate series with elongate and large plates, formed by two lateral plate series, similar in size; median plates formed by four to five irregular plate series reaching anal shield (Fig. 4a). Lateral side of body entirely covered by plates; mid-dorsal plates poorly developed, reaching middle of dorsal-fin base; median plates not interrupted in median portion of body, but with 2 or 3 plates not perforated before end of series; mid-ventral plates exceed end of anal-fin base.

Parts of head osteology presented in Fig. 4b. Tip of snout formed by a single almost square rostral plate (r). Nasal (n) almost rectangular, forming anterior medial nostril margin in contact posteriorly with frontals (f), and anterior and lateral margins contacting pre-nasals (pn). Lateral surface of head formed by three posterior rostrum plates (pr1–pr3) similar in size. Complete infraorbital plate series, present just below posterior rostrum series, composed of five plates (io1–io5), fourth infraorbital expanded ventrally, all associated with latero-sensory canal system; first infraorbital (io1) largest and fifth smallest (io5). Preopercle (pop) present just under fifth infraorbital (io5); an elongated bone, covered by latero-sensory canal. Subocular cheek plates (cp1–cp2) present above preopercle plate (pop). Top of head composed of compound pterotic-supracleithrum (cpt), supraoccipital (soc), prefrontal (pf), frontal (f), and sphenotic (sp); cpt covered with fenestrae randomly distributed and with different sizes and shapes.

Color in alcohol. Large inconspicuous brown lateral stripe extending from tip of snout through inferior orbit to end of caudal peduncle Fig. 5a, e (very weak in some specimens, such as holotype Fig. 5c). Body ground color brown on dorsum, yellowish on ven-

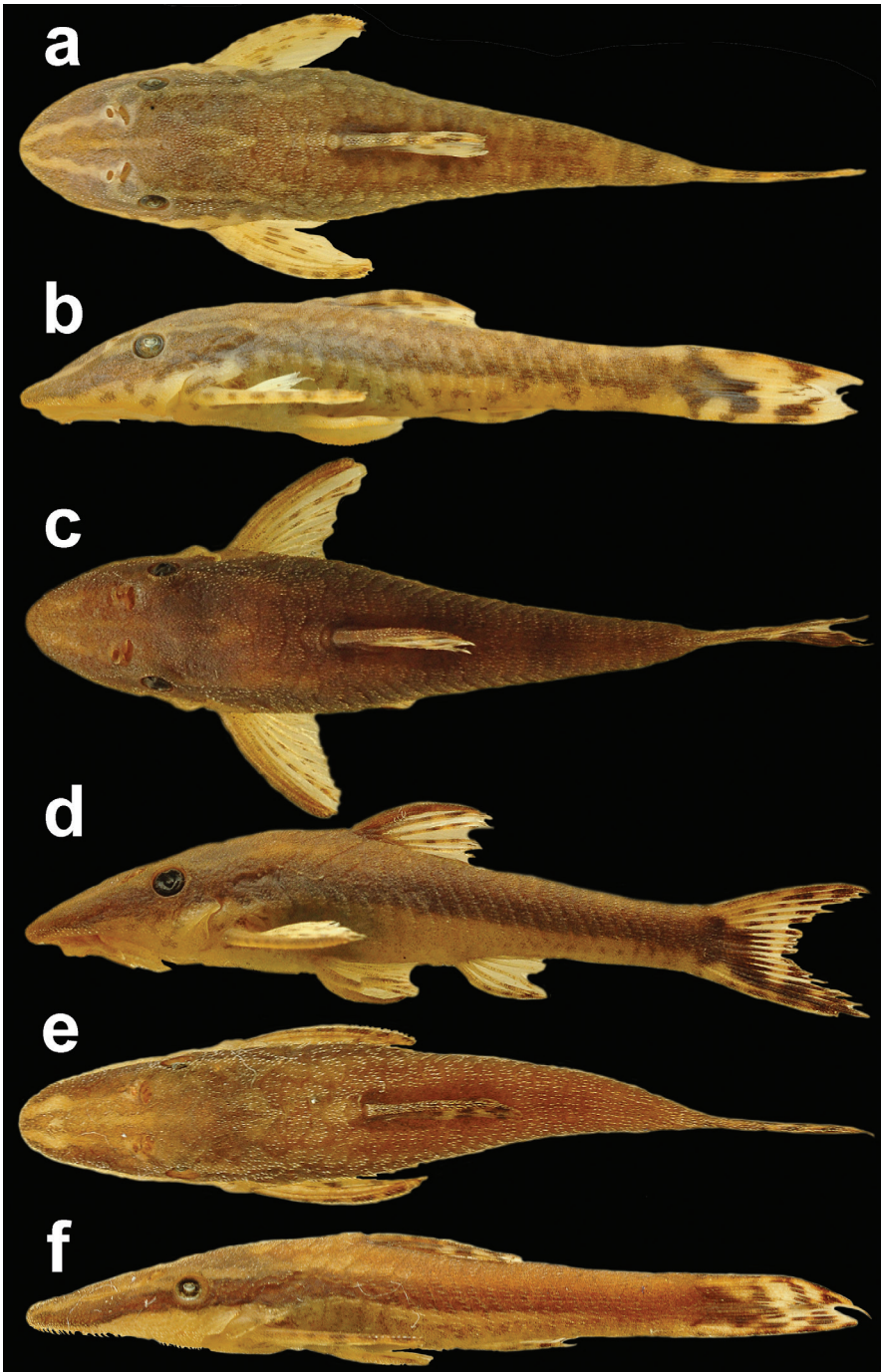


Figure 5. Variation in external morphology and coloration of *H. acuen*: **a, b** LBP 16279, 26.86 mm SL, affluent of rio Culuene, municipality of Gaúcha do Norte **c, d** MZUSP 115350, 25.9 mm SL, holotype, affluent of rio Toguro, municipality of Querência **e** (f) LBP16275, 21.67 mm SL, affluent of rio Feio, municipality of Querência.



Figure 6. *Hisonotus acuen*, LBP 16284, live specimen, from affluent of rio Toguro, Querência, Mato Grosso State, Brazil. Photo: M. Taylor.

tral region under lateral stripe. Some specimens with dark saddle on mid-ventral to ventral portion of body (Fig. 5b). Dorsal, pectoral, pelvic and anal fins with brown dots on rays, varying in concentration of chromatophores from one individual to another; inter-radial membranes hyaline. Caudal fin hyaline, except for dark blotch on origin of rays, and dark brown chromatophores largely concentrated on rays near lower caudal spine (Fig. 5d). In some specimens, chromatophores forming two dark bands on middle of rays (Fig. 5b, e).

Color in life. Similar to pattern described for alcohol individuals, but with ground color light brown (Fig. 6).

Sexual dimorphism. Males bear a papilla posterior to urogenital opening and present the pelvic-fin unbranched ray with dermal flap along its dorsal surface. Both characters are absent in females.

Distribution. *Hisonotus acuen* is known from small to median-sized streams of the upper rio Xingu basin, Mato Grosso State in Brazil (Fig. 7a).

Habitat. *Hisonotus acuen* was collected on flat areas in creeks of headwaters of the rio Xingu basin in places of shallow clear waters with low current. The fishes are found associated with vegetation that covers the bottom and the border of the headwaters (Fig. 7b).

Etymology. The specific name “acuen” is in reference to the Xavante indigenous peoples, who in anthropological literature are known as “acuen”. These people are constituted by the natives inhabiting the east of the Mato Grosso State, living in the margins of the rivers Culuene, Xingu, Mortes and Araguaia.

Discussion

The new species has a functional V-shaped spinelet (Fig. 2a–f). Carvalho and Datovo (2012) first reported this structure in *H. bockmanni*, *H. chromodontus*, *H. insperatus* and *H. luteofrenatus*. Subsequently, Roxo et al. (2014) reported this character in *H. oliveirai* and *H. paresi*. The functional V-shaped spinelet (Fig. 2a–f) is a putative apomorphic character within *Hisonotus*, and may distinguish a monophyletic group within the genus (Carvalho and Datovo 2012). However, Martins et al. (2014) have a different interpretation, in which the spinelet in *H. chromodontus*, *H. luteofrenatus* and

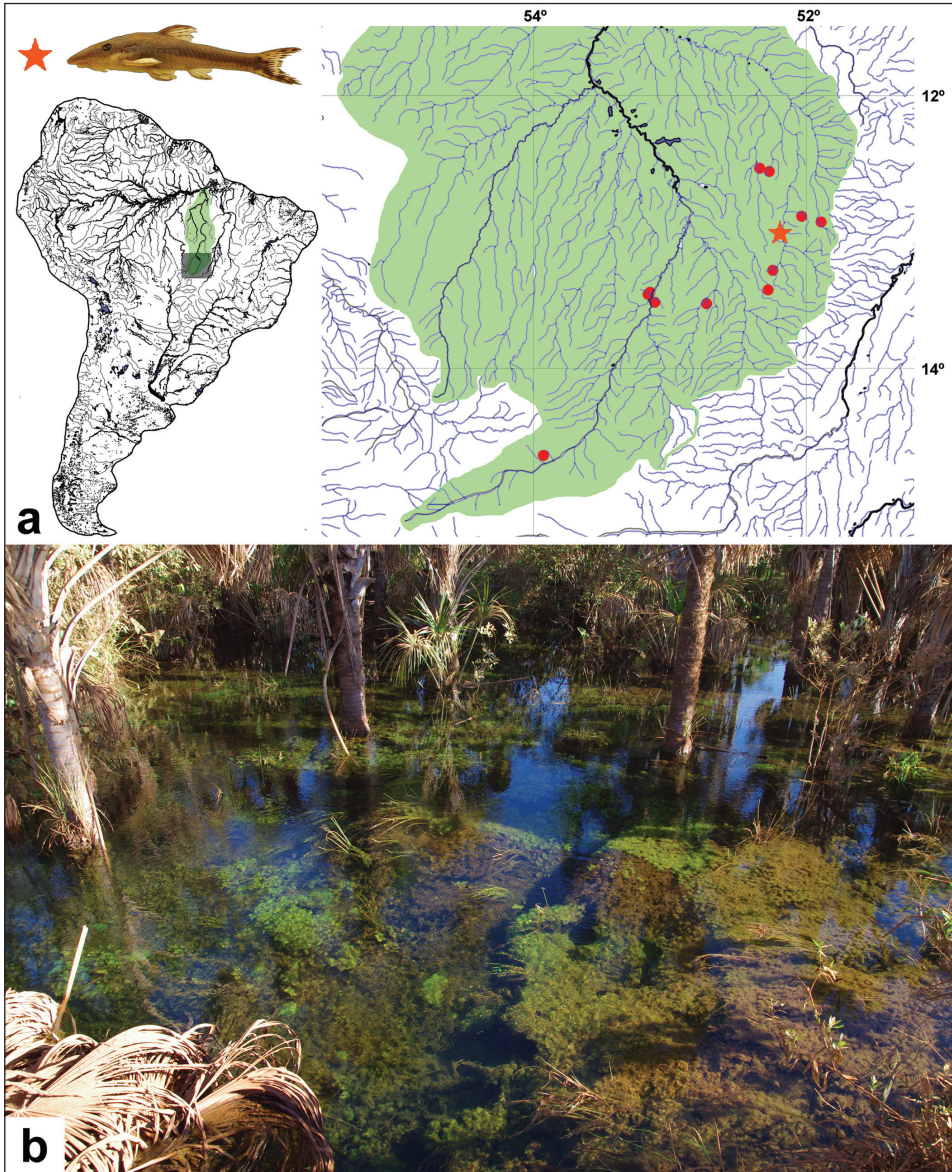


Figure 7. a Map of the distribution of *H. acuen*. Red star = holotype locality, affluent of rio Toguro. Red circles = paratype localities, affluent of rio Culuene, affluent of rio Suiá-Missu, affluent of rio Feio, córrego Xavante, rio Coronel Vanick. All are tributaries of rio Xingu, Mato Grosso State, Brazil **b** Affluent of rio Feio, municipality of Querência, 12°33'20"S, 52°16'16"W, habitat where the specimens of *H. acuen* were found. Photo: M. Taylor.

H. piracanjuba is reduced, and the locking mechanism is not functional (Martins et al. 2014, Fig. 11A), the same character state found in *H. armatus*, *H. depressicauda*, *H. francirochai* and *H. notatus*. Martins et al. (2014) also suggested that in *H. insperatus*

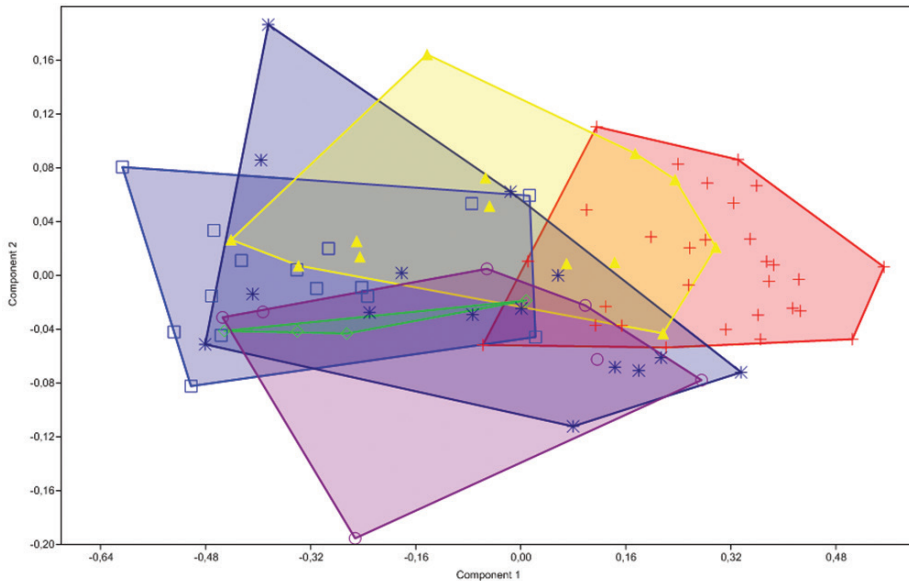


Figure 8. Scatter plot of Principal Component Analysis (PCA) of six allopatric populations of *H. acuen* ($n = 83$) indicating the presence of continuous external morphology variation. Purple circle = affluent of rio Toguro; Red cross = affluent of rio Culuene; Dark blue star = affluent of rio Suiá-Missu; Blue square = affluent of rio Feio; Yellow triangle = córrego Xavante; Green diamonds = rio Coronel Vanick. All are tributaries of rio Xingu, Mato Grosso State in Brazil.

the spinelet is absent (Martins et al. 2014, Fig. 11b). However, in our interpretation, *H. acuen*, *H. chromodontus*, *H. bockmanni*, *H. insperatus*, *H. luteofrenatus*, *H. oliveirai*, *H. parsi*, and *H. piracanjuba* exhibit a functional V-shaped spinelet, which is not present in *H. depressicauda* and *H. notatus* (Fig. 2). Therefore, despite the fact that the genus *Hisonotus* may not represent a monophyletic unit, we include *H. acuen* within *Hisonotus* pending a formal phylogenetic analysis of Hypoptopomatinae, and the species-level composition is established.

Hisonotus acuen exhibits a large amount of variation in external body proportions across its range (Fig. 5), especially in body depth at dorsal-fin origin (13.5–22.8% of SL), snout length (34.2–57.2% of HL), and abdomen length 10.2–24.4% of SL). This variation is partly distributed within populations, and partly between populations, and is not strongly correlated with body size. We performed a PCA to evaluate the morphometric variation within this new species. We compared the morphometric data of six populations found in different tributaries of the rio Xingu, and our results suggest that the range in morphology has a continuous distribution. The lack of phenotypic discontinuities among populations suggests they are not different species (Fig. 8). Additionally, we found that the genetic variation of the Cytochrome Oxidase I (COI) gene within the populations of *H. acuen* is 1%, and that variation among closely related congeners (i.e. *H. chromodontus*, *H. insperatus*, *H. oliveirai*, *H. parsi* and *H. piracanjuba*) is more than 17% (see Table 3 and Fig. 9; sequences can



Table 3. Genetic distance (and standard deviation) between *Hisonotus* species and specimens of the same species (main diagonal). This analysis was performed using Kimura 2-parameter substitution model, Gamma distribution and 1,000 bootstrap pseudoreplicates.

	1	2	3	4	5	6	7
1. <i>H. acuen</i>	1.0±0.2						
2. <i>H. paresi</i>	17.0%±2.3	0±0					
3. <i>H. piracanjuba</i>	21.2%±2.6	6.8%±1.3	0±0				
4. <i>H. chromodontus</i>	19.3%±2.5	19.5%±2.6	20.0%±2.6	0±0			
5. <i>H. notatus</i>	20.5%±2.8	19.0%±2.7	18.5%±2.4	22.3%±2.9	0±0		
6. <i>H. insperatus</i>	21.4%±2.7	8.0%±1.4	5.1%±0.9	21.6%±2.7	16.1%±2.1	0.1±0.1	
7. <i>H. oliveirai</i>	20.2%±2.5	6.8%±1.2	4.1%±0.9	21.2%±2.7	18.1%±2.3	3.0%±0.7	0.6±0.3

be downloaded from GenBank using the accession numbers provided in Methods). Therefore, the available morphological and molecular data support the recognition of the individuals inhabiting the six localities of rio Xingu and representing different populations as a single species.

The new species *H. acuen* is the first described species of *Hisonotus* from the rio Xingu basin, and is externally very similar to *H. chromodontus*, a species from the rio Tapajos basin. The coloration of the caudal fin and the tip of the teeth distinguish these species that also are very different genetically (i.e. 19.3% of genetic divergence; Table 3 and Fig. 9). Britski and Garavello (2003) discussed the coloration of the teeth of *H. chromodontus*, reporting that in more than one hundred specimens examined, varying from 12.0 to 32.2 mm SL, all tooth-tips have a reddish-brown color. We analyzed more than one hundred specimens of *H. chromodontus* from the museum collections of LBP and NUP, and found the same reddish-brown tooth tips. This tooth features appears to be unique within the genus *Hisonotus*. A very similar external morphology, as well as the presence of the functional V-shaped spinelet among *H. acuen* and *H. chromodontus*, could suggest a close relationship between these species.

Comparative material

All from Brazil, except when stated otherwise: *Hisonotus aky* (Azpelicueta, Casciotta, Almirón & Koerber, 2004): MHNG 2643.039, 2, 33.1–34.2 mm SL, paratypes, arroio Fortaleza, Argentina; *Hisonotus bocaiúva* Roxo, Silva, Oliveira & Zawadzki, 2013: MZUSP 112204, male, 24.2 mm SL, holotype, córrego Cachoeira, Bocaiúva, Minas Gerais; LBP 9817, 9, 3 c&s, 18.3–23.2 mm SL, paratypes, córrego Cachoeira, Bocaiúva, Minas Gerais; *Hisonotus carreiro* Carvalho & Reis, 2011: MCP 40943, 3, 33.6–35.8 mm SL, arroio Guabiju, Guabiju, Rio Grande do Sul; *Hisonotus charrua* Almirón, Azpelicueta, Casciotta & Litz, 2006: LBP 4861, 1, 35.9 mm SL, arroio Guaviyú, Artigas, Uruguay; MHNG 2650.051, 1, 34.2 mm SL, paratype, arroio Aspinillar, Uruguay; *Hisonotus chromodontus* Britski & Garavello, 2007: LBP 7964, 25, 24.0–28.3 mm SL, 3 females c&s, 26.5–28.9 mm SL, 1 male c&s 24.9 mm SL, rio dos

Patos, Nova Mutum, Mato Grosso; LBP 7974, 26, 17.7–24.8 mm SL, rio dos Patos, Nova Mutum, Mato Grosso; LBP 12278, 2, 26.7–28.7 mm SL, 1 unsexed c&s, 26.7 mm SL, rio Sumidouro, Tangará da Serra, Mato Grosso; MZUSP 45355, 25.9 mm SL, holotype, affluent of rio Preto, Diamantino, Mato Grosso; MZUSP 70758, 7, 19.4–23.9 mm SL, paratype, riacho Loanda, Sinop, Mato Grosso; NUP 10924, 24, 19.5–31.5 mm SL, rio Preto, Diamantino, Mato Grosso; *Hisonotus depressicauda* (Miranda Ribeiro, 1918): MZUSP 5383, 24.4 mm SL, paralectotype, Sorocaba; LBP 17474, 5 c&s, 18.1–24.0 mm SL, rio Araquá, Botucatu, São Paulo; *Hisonotus francirochai* (Ihering, 1928): LBP 13923, 22, 25.7–35.7 mm SL, córrego sem nome, Capitanga, Minas Gerais; MZUSP 3258, 29.4 mm SL, lectotype, rio Grande, São Paulo; *Hisonotus heterogaster* Carvalho & Reis, 2011: LBP 3335, 39, 20.8–30.1 mm SL, arroio sem nome, rio Grande, Rio Grande do Sul; *Hisonotus insperatus* Britski & Garavello, 2003: LBP 1299, 3, 23.5–29.6 mm SL, 1 female c&s, 24.8 mm SL, rio Araquá, Botucatu, São Paulo; LBP 1316, 2, 24.1–27.4 mm SL, 1 female c&s, 24.7 mm SL, 1 male c&s, 23.9 mm SL, rio Araquá, Botucatu, São Paulo; LBP 1344, 2, 22.9–24.9 mm SL, rio Araquá, Botucatu, São Paulo; LBP 1373, 1, 25.8 mm SL, rio Araquá, Botucatu, São Paulo; LBP 1405, 2, 22.2–27.3 mm SL, rio Araquá, Botucatu, São Paulo; LBP 4699, 17, 19.6–26.9 mm SL, 4 females c&s, 20.3–26.8 mm SL, 3 males c&s, 24.3–26.1 mm SL, ribeirão Cubatão, Marapoama, São Paulo; LBP 4945, 5, 27.3–28.5 mm SL, 2 females c&s, 28.2–29.9 mm SL, Botucatu, São Paulo; LBP 6770, 5, 25.1–28.2 mm SL, 3 females c&s, 20.0–27.0 mm SL, ribeirão Cubatão, Marapoama, São Paulo; LBP 13336, 1 female c&s, 26.0 mm SL, rio Capivara, Botucatu, São Paulo; LBP 13337, 2 females c&s, 27.4–28.6 mm SL, rio Araquá, Botucatu, São Paulo; MZUSP 22826, 1, 25.4 mm SL, paratype, córrego Água Tirada, Três Lagoas, Mato Grosso; MZUSP 24832, 1, 23.8 mm SL, paratype, rio Corumbataí, Corumbataí, São Paulo; MZUSP 78957, 29.6 mm SL, holotype, rio Capivara, Botucatu, São Paulo; MZUSP 78960, 31, 12.6–26.0 mm SL, paratypes, 5 c&s, 22.7–24.7 mm SL, rio Pardo, Botucatu, São Paulo; MZUSP 78965, 10, 15.6–28.6 mm SL, paratypes, 3 c&s, not measured, rio Araquá, Botucatu, São Paulo; MZUSP 78968, 5, 24.1–27.3 mm SL, paratypes, córrego da Figueira, Lins, São Paulo; *Hisonotus iota* Carvalho & Reis, 2009: LBP 13072, 5, 32.3–33.0 mm SL, rio Chapecó, Coronel Freitas, Santa Catarina; *Hisonotus laevior* Cope, 1894: LBP 3377, 1, 25.2 mm SL, arroio dos Corrientes, Pelotas, Rio Grande do Sul; LBP 6037, 8, 33.4–47.0 mm SL, rio Maquiné, Osório, Rio Grande do Sul; LBP 13187, 7, 19.4–45.8 mm SL, córrego sem nome, Camaquã, Rio Grande do Sul; *Hisonotus leucofrenatus* (Miranda Ribeiro, 1908): LBP 2085, 7, 38.3–50.6 mm SL, rio Sagrado, Morretes, Paraná; LBP 6837, 36, 35.1–43.5 mm SL, rio Fau, Miracatu, São Paulo; *Hisonotus leucophrys* Carvalho & Reis, 2009: LBP 13065, 6, 17.2–33.6 mm SL, rio Ariranhas, Xavantina, Santa Catarina; LBP 13073, 1, 36.8 mm SL, rio Guarita, Palmitinho, Rio Grande do Sul; *Hisonotus luteofrenatus* Britski & Garavello, 2007: MZUSP 62593, 28.6 mm SL, holotype, córrego Loanda, Cláudia, Mato Grosso; MZUSP 62594, 8, 22.4–30.5 mm SL, paratype, riacho Selma, Sinop, Mato Grosso; MZUSP 95940, 3, 26.1–28.5 mm SL, affluent of rio Teles Pires, Itaúba, Mato Grosso; *Hisonotus megaloplax* Carvalho & Reis, 2009: LBP 13108, 6, 36.4–37.8 mm SL, cór-

rego sem nome, Saldanha Marinho, Rio Grande do Sul; *Hisonotus montanus* Carvalho & Reis, 2009: LBP 13051, 3, 26.4–27.2 mm SL, rio Goiabeiras, Vargem, Santa Catarina; LBP 13055, 5, 24.8–31.9 mm SL, rio Canoas, Vargem, Santa Catarina; *Hisonotus nigricauda* (Boulenger, 1891): LBP579, 16, 34.1–40.1 mm SL, rio Guaíba, Eldorado do Sul, Rio Grande do Sul; *Hisonotus notatus* Eigenmann & Eigenmann, 1889: LBP 3472, 20, 21.0–34.3 mm SL, 2 males c&s 25.8–26.5 mm SL, 1 female c&s, 25.0 mm SL, rio Aduelas, Macaé, Rio de Janeiro; LBP 10742, 25, 24.4–43.3 mm SL, rio Macabu, Conceição de Macabu, Rio de Janeiro; *Hisonotus oliveirai* Roxo, Zawadzki & Troy, 2014: MZUSP 115061, female, 26.4 mm SL, holotype, ribeirão Cambira, affluent of rio Ivaí, Cambira, Paraná; LBP 13332, 1 male, 23.2 mm SL, 1 unsexed c&s, 23.7 mm SL, paratype, rio Mourão, rio Ivaí basin, Campo Mourão, LBP 17578, 3 females, 27.7–30.4 mm SL, 2 males, 25.4–26.1 mm SL, paratypes, rio Mourão, rio Ivaí basin, boundary between Engenheiro Beltrão and Quinta do Sol; NUP 3578, 7 females, 27.8–28.1 mm SL, 8 males, 24.7–26.8 mm SL, 1 female c&s, 27.6 mm SL, 1 male c&s, 25.5 mm SL, paratypes, ribeirão Salto Grande, rio Ivaí basin, Maria Helena; *Hisonotus parsi* Roxo, Zawadzki & Troy, 2014: MZUSP 115062, female, 26.2 mm SL, holotype, riacho Águas Claras, affluent of rio Sepotuba, Santo Afonso; LBP 13351, 9, 14.7–24.3 mm SL, paratype, riacho Águas Claras, Santo Afonso; LBP 13352, 1, 23.7 mm SL, paratype, riacho Águas Claras, Santo Afonso; NUP 10928, 2 males, 23.2–24.2 mm SL, paratype, 2 c&s, 23.6–24.2 mm SL, riacho Águas Claras, affluente of rio Sepotuba, Santo Afonso; NUP 10976, 3 unsexed, 16.7–20.5 mm SL, paratype, riacho São Jorge, Santo Afonso; *Hisonotus piracanjuba* Martins & Langeani, 2012: LBP 17256, 9, 17.2–26.3 mm SL, 1, c&s 27.1 mm SL, córrego sem nome, Morrinhos, Goiás; NUP 5059, 1, 24.7 mm SL, córrego Posse, Anápolis, Goiás; NUP 10979, 3, 21.4–21.8 mm SL, ribeirão Bocaina, Piracanjuba, Goiás; *Hisonotus prata* Carvalho & Reis, 2011: MCP 40492, 18, 19.5–33.2 mm SL, rio da Prata, Nova Prata, Rio Grande do Sul; LBP 9918, 14, 21.7–32.6 mm SL, Laguna dos Patos system, Nova Prata, Rio Grande do Sul; *Hisonotus ringueleti* Aquino, Schaefer & Miquelarena, 2001: FMNH 108806, 2, 25.7–32.2 mm SL, rio Quaraí basin, Uruguay; LBP 13148, 1, 24.5 mm SL, arroio Putiá, Uruguiana, Rio Grande do Sul; *Hisonotus* sp.: LBP 8276, 1 c&s, 25.6 mm SL, rio Verde Grande, Jaíba, Minas Gerais; *Microlepidogaster arachas* Martins, Calegari & Langeani, 2013: LBP 10882, 3, 22.8–35.3 mm SL, rio Paraná basin, Araxás, Minas Gerais; *Microlepidogaster dimorpha* Martins & Langeani, 2011: LBP 10683, 2, 28.8–35.6 mm SL; rio Paraná basin, Uberaba, Minas Gerais; *Otothyris travassosi* Garavello, Britski & Schaefer, 1998: LBP 1971, 13, 14.0–27.2 mm SL; coastal drainage, Canavieiras, Bahia; *Otothyropsis marapoama* Ribeiro, Carvalho & Melo, 2005: LBP 4698, 6, 23.9–36.3 mm SL; rio Tietê basin, Marapoama, São Paulo; *Parotocinclus aripuanensis* Garavello, 1988: LBP 10981, 33, 15.63–18.47 mm SL, rio Lageado, Guajará Mirim, Rondônia; *Parotocinclus* aff. *spilurus* (Fowler, 1941): LBP 5624, 21.5–22.0 mm SL, rio Maravilha, Balsas, Maranhão; *Parotocinclus* cf. *bahiensis* (Miranda Ribeiro, 1918): LBP 7182, 3, 27.9–35.6 mm SL; rio Paraguaçu basin, Lençóis, Bahia; *Parotocinclus maculicauda* (Steindachner, 1877): LBP 2869, 15, 20.2–44.7 mm SL, rio Ribeira do Iguape basin, Miracatu, São Paulo; *Parotocinclus prata* Ribeiro,

Melo & Pereira, 2002: LIRP 1136, 38, 19.8–41.9 mm SL; rio São Francisco basin, Presidente Olegário, Minas Gerais; *Parotocinclus* sp.: LBP 1572, 3, 19.0–24.0 mm SL, ribeirão Ínsula, Barra do Garça, Mato Grosso; LBP 2414, 19, 16.7–20.8 mm SL, 1 c&s 23.6 mm SL, córrego Fundo, Barra do Garça, Mato Grosso.

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