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A new species of the genus *Henricia* (Asteroidea, Spinulosida, Echinasteridae) from South Korea

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Abstract

A new species of the genus *Henricia* Gray, 1840 that belongs to the family Echinasteridae is described from South Korea. *Henricia epiphysialis* **sp. nov.** has epiphyseal ossicles at the ends of the abactinal and lateral plates, and the abactinal and lateral spines form a hooked crown. The partial sequence of the mitochondrial *COXI* gene (537 bp) of *H. epiphysialis* **sp. nov.** was obtained, and the new species was morphologically and genetically compared with other related *Henricia* species.

Keywords

Distribution, DNA barcoding, *Henricia epiphysialis* **sp. nov.**, morphology, taxonomy

Introduction

Echinasteridae Verrill, 1867 is the only family that belongs to the order Spinulosida Perrier, 1884. This family comprises eight accepted genera: *Aleutihenricia* Clark & Jewett, 2010; *Dictyaster* Wood-Mason & Alcock, 1891; *Echinaster* Müller & Troschel, 1840; *Henricia* Gray, 1840; *Metrodira* Gray, 1840; *Odontohenricia* Rowe & Albertson, 1988; *Plectaster* Sladen, 1889; and *Rhopiella* Fisher, 1940 (Mah 2019). Most of the species are found in genera *Echinaster* and *Henricia*.

A total of 11 species that belong to *Aleutihenricia* or *Henricia* have been reported in South Korea: *Aleutihenricia beringiana* Djakonov, 1950 and 10 *Henricia* species, namely, *Henricia anomala* Hayashi, 1973; *Henricia elachys* Clark & Jewett, 2010; *Henricia leviuscula* Stimpson, 1857; *Henricia nipponica* Uchida, 1928; *Henricia ohshimai* Hayashi, 1935; *Henricia pachyderma* Hayashi, 1940; *Henricia pacifica* Hayashi, 1940; *Henricia regularis* Hayashi, 1940; *Henricia reniossa* Hayashi, 1940; and *Henricia sanguinolenta* O.F. Müller, 1776 (Shin 2010; Shin and Ubagan 2015a, b; Ubagan and Shin 2016, 2019a, b, c; Taekjun and Shin 2020). Most species recorded in South Korea including *H. leviuscula*, *H. nipponica*, *H. pachyderma*, *H. regularis*, *H. reniossa*, and *H. sanguinolenta*, are distributed in the East Sea. *Henricia* species can be distinguished by the ratio of arm to disk, shape and number of abactinal spines, and shape of the skeletal plates.

In DNA barcoding, sequence variation in a 658 bp region of the mitochondrial cytochrome *c* oxidase subunit I (*COXI*) gene is used for specimen identification and species discovery (Hebert et al. 2003). An integrative approach to taxonomy (i.e., using morphological characteristics from preserved specimens as well as one to several genes) has become necessary for assessing species diversity and species boundaries (Puillandre et al. 2012).

In this study, we identified a new species that belongs to the genus *Henricia* collected from waters adjacent to the East Sea, South Korea, and performed detailed morphological and molecular mitochondrial sequence analyses. This paper aims to extend the taxonomical insights to *Henricia* species in South Korea by providing a complete description of this new species.

Materials and methods

In May and December 2014, sea stars were collected from the East Sea in South Korea by using fishing nets (Fig. 1). The collected specimens were preserved in 95% ethanol and deposited at the National Institute of Biological Resources (NIBR) and Marine Echinoderm Resource Bank of Korea (MERBK), South Korea. The external features of the specimens were observed using a stereomicroscope, and the specimens were identified on the basis of morphological characteristics such as the size of the disk, R/r ratio (R: length of arm; r: radius of the disk), size of the upper and proximal portions of arms, number of abactinal spines, shape of abactinal and actinal skeletons, and number of adambulacral spines. For observing the detailed structures of the specimens such as the shape of the spines and skeletal plates, sodium hypochlorite (5.25% solution) was applied carefully to dissolve the skin (Shin 2010). Then, the specimens were washed with water and observed using the stereomicroscope. The important morphological characteristics of the specimens were photographed using a scanning electron microscope (JEOL JSM-6510), stereomicroscope (Nikon SMZ1000), and digital camera (Nikon D7000). Abbreviations for the measurements were those used by Shin and Ubagan (2015a, b).

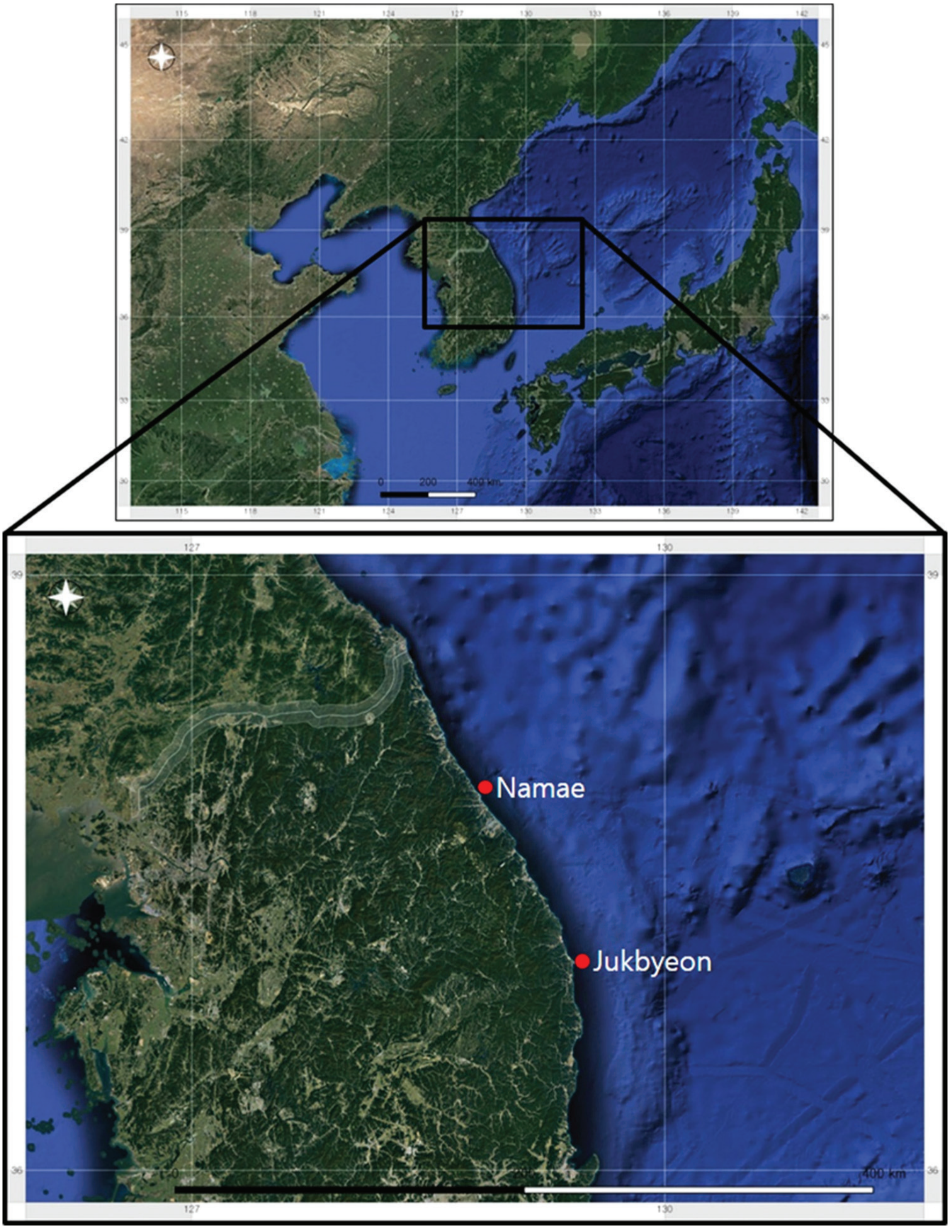

Figure 1. Map of Korea showing the collection sites of *Henricia epiphysialis* sp. nov. in the East Sea, South Korea.

Total genomic DNA was isolated from ethanol-preserved tube feet tissue by using a DNeasy blood and tissue DNA isolation kit (Qiagen), according to the manufacturer instructions. The genomic DNA quality and concentration were determined using a Nanodrop ND-1000 spectrophotometer (Thermo Fisher Scientific). All

Table 1. List of *Henricia* species and GenBank accession numbers of *COX1* gene used in this study.

Species	GenBank No.	Dataset		References
		189 bp	537 bp	
H. compacta (Sladen, 1889)	KT268147	+		Lopes et al. 2016
H. epiphysialis sp. nov.	MT086587	+	+	present study
H. hayashii Djakonov, 1961	LC336732	+	+	Wakita et al. 2019
H. hedingi Madsen, 1987	KY853274	+		Knott et al. 2018
H. kinkasana Hayashi, 1940	LC336731	+	+	Wakita et al. 2019
H. leviuscula (Stimpson, 1857)	MK947912	+	+	Lee and Shin, 2019
H. lisa Clark, 1949	KY853275	+	+	Knott et al. 2018
H. nipponica Uchida, 1928	LC336733	+	+	Wakita et al. 2019
H. obesa (Sladen, 1889)	KT268148	+		Lopes et al. 2016
H. oculata (Pennant, 1777)	KT268151	+	+	Knott et al. 2018
H. ohshimai Hayashi, 1935	LC336735	+	+	Wakita et al. 2019
H. ohshimai Hayashi, 1935	LC336736	+	+	Wakita et al. 2019
H. pachyderma Hayashi, 1940	MT079801	+	+	Lee and Shin, 2020
H. perforata (Müller, 1776)	KY853302	+	+	Knott et al. 2018
H. pertusa (Müller, 1776)	KY853286	+	+	Knott et al. 2018
H. regularis Hayashi, 1940	LC336739	+	+	Wakita et al. 2019
H. reniossa Hayashi, 1940	LC336740	+	+	Wakita et al. 2019
H. reticulata Hayashi, 1940	LC336737	+	+	Wakita et al. 2019
H. sanguinolenta (Müller, 1776)	HM542200	+		Lopes et al. 2016
H. sanguinolenta (Müller, 1776)	KY853253	+	+	Knott et al. 2018
H. spongiosa (Fabricius, 1780)	KY853268	+	+	Knott et al. 2018
H. tumida Verrill, 1909	LC336747	+	+	Wakita et al. 2019
Henricia sp. 1	LC336744	+	+	Wakita et al. 2019
Henricia sp. 2	LC336742	+	+	Wakita et al. 2019
Henricia sp. 3	LC336743	+	+	Wakita et al. 2019
Henricia sp. 4	LC336741	+	+	Wakita et al. 2019
Henricia sp. 5	LC336738	+	+	Wakita et al. 2019
Henricia sp. 6	LC336745	+	+	Wakita et al. 2019
Henricia sp. 7	LC336746	+	+	Wakita et al. 2019
Henricia sp. 8	LC336730	+	+	Wakita et al. 2019
Henricia sp. 9	KY853310	+	+	Knott et al. 2018
Echinaster brasiliensis Müller & Troschel, 1842	MG636999	+	+	Seixas et al. 2018
E. callosus Marenzeller, 1895	KT268121	+		Lopes et al. 2016
E. luzonicus (Gray, 1840)	KT268137	+		Lopes et al. 2016
E. sepositus (Retzius, 1783)	LC336729	+	+	Wakita et al. 2019

genomic DNA samples were stored at –20 °C until further use. The partial sequence of the mitochondrial *COI* gene (658 bp) was amplified using a pair of primers, LCOech1aF1 (Layton et al. 2016) and HCO2198 (Folmer et al. 1994). PCR was performed using a 25 µL reaction mixture containing 2.5 µL of 10× Ex Taq Buffer containing 20 mM $MgCl_2$ (Clontech), 1 µL of 2.5 mM dNTPs (Clontech), 1 µL of each primer at 10pmol, 1.5 µL of the template DNA, 0.3 µL of 5 U/µL Taq polymerase (Clontech), and 17.7 µL of distilled water. The PCR conditions were as follows: initial denaturation at 95 °C/3 min, followed by 35 cycles of denaturation at 95 °C/30 s, annealing at 52 °C/90 s, extension at 72 °C/90 s, and a final extension at 72 °C/7 min. The PCR product quality was determined using electrophoresis with a 1.5% agarose gel stained with ethidium bromide. The PCR products were directly sequenced in both directions by using ABI Big Dye Terminator kits (Applied Biosystems) and ABI 3730XL DNA Analyzer. To construct a neighbor-joining (NJ) tree, *COX1* sequences (189 and 537 bp) from the *Henricia* species dataset, including one sequence of the new species (GenBank accession No. MT086587), were used (Table

1). Four species of genus *Echinaster* were used as outgroups (Table 1). Pairwise distances were calculated using MEGA 7.0 and the Kimura-2-parameter model (Kumar et al. 2016). The gaps and missing data were removed, and the bootstrap analysis was performed with 1000 replicates.

Taxonomic results

Phylum Echinodermata Bruguière, 1791
Class Asteroidea de Blainville, 1830
Order Spinulosida Perrier, 1884
Family Echinasteridae Verrill, 1867
Genus *Henricia* Gray, 1840

***Henricia epiphysialis* sp. nov.**
http://zoobank.org/E50768C6-B625-43ED-82E1-6D581E38975B
Figs 2–4

Material examined. *Holotype*: South Korea • 1 specimen; waters adjacent to Namae, 37°55'57.31"N, 128°48'45.58"E; 40 m; 28 May 2014; S. Shin and T. Lee; fishing net; MERBK–A–1255. ***Paratypes*:** South Korea • 1 specimen; waters adjacent to Jukbyeon, 37°3'32.49"N, 129°26'14.57"E; 100 m; 19 Dec. 2014; S. Shin and T. Lee; fishing net; NIBRIV0000837785. 1 specimen; waters adjacent to Namae, 37°55'57.31"N, 128°48'45.58"E; 40 m; 28 May 2014; S. Shin and T. Lee; fishing net; MERBK–A–1256.

Diagnosis. Regular size, R/r = 4.9–5.4, abactinal plates crowded with 11–40 spines, abactinal and lateral spines forming distinct hooked crown, epiphyseal ossicles formed at ends of abactinal and lateral plates, one to three papulae, marginal and ventrolateral series distinguishable, adambulacral plates bearing 10–14 slender spines.

Description. *Holotype.* (Figs 2–4) Size. R = 51 mm, r = 10 mm, R/r = 5.1.

Arms five, semi-cylindrical, gradually tapering to tips (Fig. 2A–B). Abactinal paxillae formed in group with evenly spaced spinulation, bearing 11–40 spines with serrated tips (Fig. 2C). Denuded abactinal spines forming hooked crown composed of nine to 11 large hook-shaped spinules enclosing nine to 12 small connected apical tips (Fig. 4A–C). Paxillae on lateral side of arms similar to abactinal paxillae (Fig. 2D). Denuded abactinal plates reniform, usually connected to end of other plate in mid convex part of plate, larger than papular areas, partially enclosing papular area on concave side of plate. Papular areas narrow, containing one to three papulae in an area. Some papular areas divided by small ossicles (Fig. 3A). Almost every skeletal plate aside from adambulacral plates was observed bearing epiphyseal ossicles at ends of plate (Fig. 3A, C, D). Madreporite circular in form, slightly elevated, bearing spines same as adjacent spines (Fig. 3B). Shape of spines on lateral side nearly similar to that of abactinal spines (Fig. 4D–F). Superomarginal, inferomarginal, and ventrolateral plates well defined forming elongated cross shape and arranged in rows show-

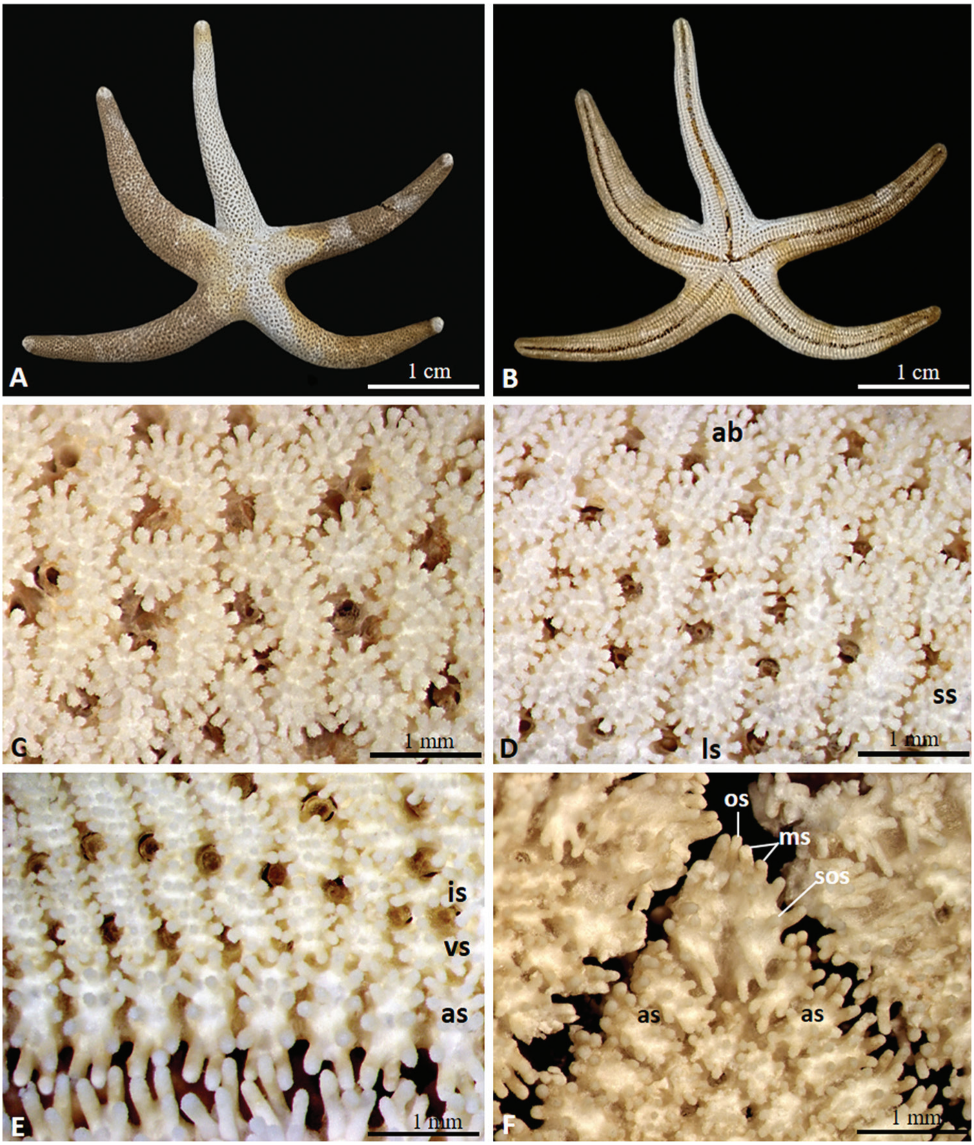

Figure 2. External characteristics of *Henricia epiphysialis* sp. nov. **A** abactinal side **B** actinal side **C** abactinal spines **D** spines on lateral side of arm **E** adambulacral spines **F** oral part. Abbreviations: **ab** abactinal side **ls** lateral side **ss** superomarginal spines **is** inferomarginal spines **vs** ventrolateral spines **as** adambulacral spines **os** oral spines **ms** marginal spines **sos** sub-oral spines.

ing consistent series (Fig. 3D). Superomarginal plates bearing 12–28 spines, bend upward toward base of arm in crescentic form, and reach tip of arm (Figs 3C–D, 4D). Intermarginal plates forming small elongated shape, extending near half of arm (Fig. 3D). Inferomarginal plates longer than superomarginal and ventrolateral plates, bearing 34–45 spines, reaching tip of arm (Figs 3C–D, 4E). Ventrolateral plates

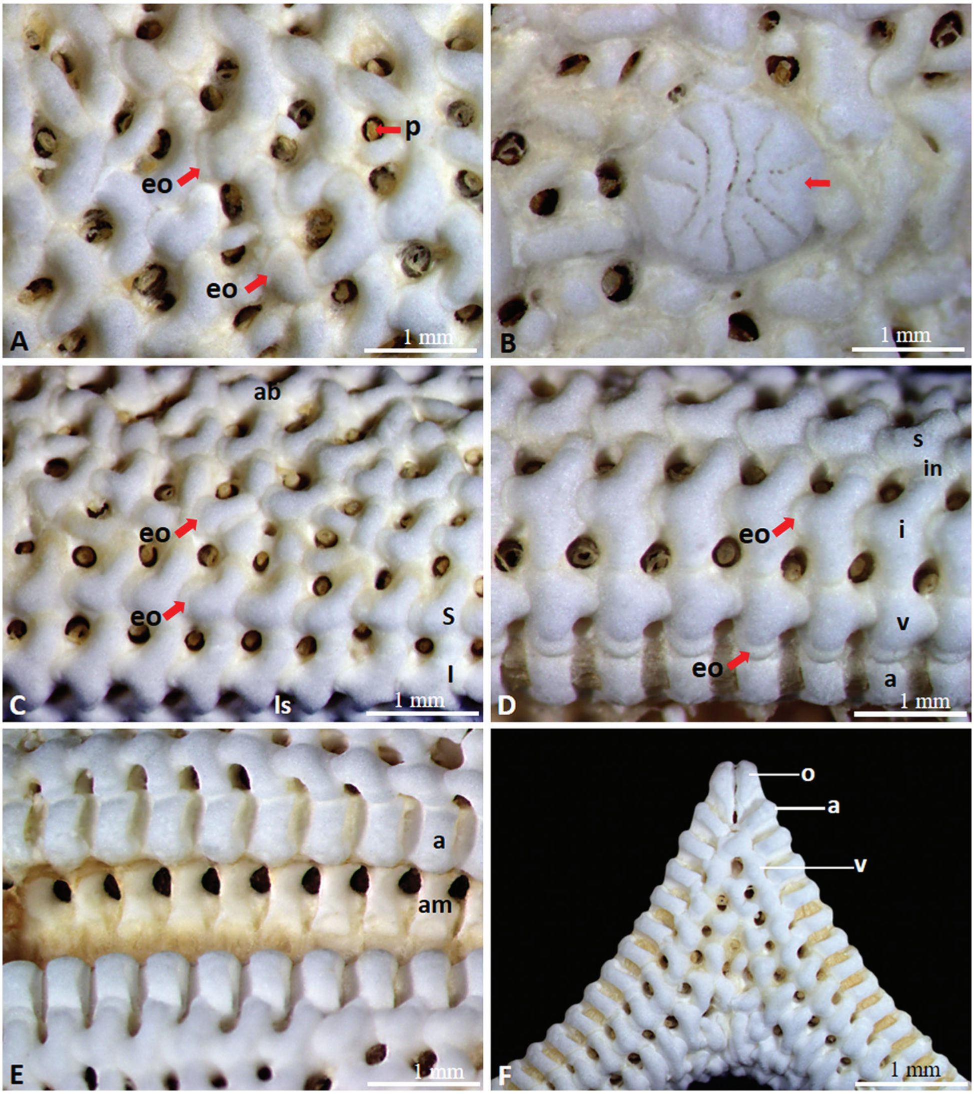

Figure 3. Denuded skeleton of *Henricia epiphysialis* sp. nov. **A** abactinal plates **B** madreporite (arrow) **C** part of abactinal and lateral side of arm **D** plates on the lateral and actinal side of arm **E** actinal plates **F** oral part. Abbreviations: **ab** abactinal side **ls** lateral side **eo** epiphyseal ossicles **p** papula **s** superomarginal plates **in** intermarginal plates **i** inferomarginal plates **v** ventrolateral plates **a** adambulacral plates **am** ambulacral plates **o** oral plates.

forming a rounded cross shape, bearing 21–25 spines, reaching near tip of arm, epiphyseal ossicles forming a knob-like connection to adambulacral plates, extending to middle part of arm (Figs 3D, 4F). Adambulacral plates forming semi-rounded shape, bearing 10–14 slender, thorny spines, arranged in two transverse series (Figs 3D–E, 4G), articulated with ambulacral plates (Fig. 3E). Furrow spine single, somewhat

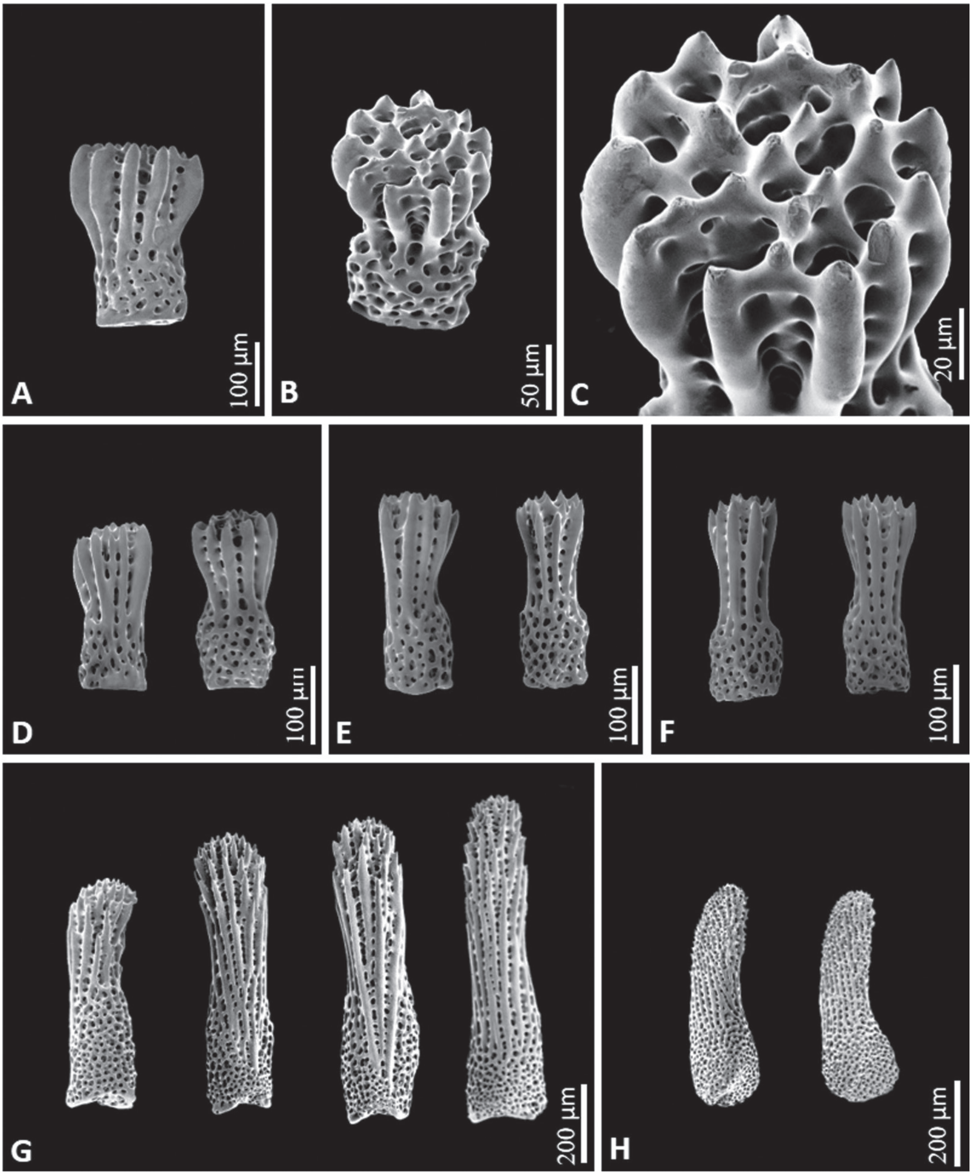

Figure 4. SEM images of the spines of *Henricia epiphysialis* sp. nov. **A, B, C** abactinal spines **D** superomarginal spines **E** inferomarginal spines **F** ventrolateral spines **G** adambulacral spines **H** furrow spines.

curved (Fig. 4H). Oral part bearing two slender, bluntly pointed oral spines, with six or seven marginal spines, and five or six sub-oral spines similar to adambulacral spines (Fig. 2F). Paired oral plates forming a slightly elongated triangular shape, articulated with first pair of adambulacral plates. Plates of inter-radial area slightly compact (Fig. 3F).

Paratypes. Size. R = 39 mm, r = 8 mm, R/r = 4.9; R = 60 mm, r = 11 mm, R/r = 5.4.

Etymology. The specific name is derived from the Latin "epiphysialis," which means the end part of a long bone.

Ecology. This species is found on hard substrates (rocky bottom) from a shallow water of a depth of 40 m to 100 m.

Distribution. South Korea (East Sea).

Molecular results

We determined the phylogenetic relationships based on two *COX1* datasets with 27 and 31 species of the genus *Henricia* respectively, including *H. epiphysialis* sp. nov., on the basis of 189 and 537 bp of the mitochondrial *COX1* gene by using the NJ method. All *Henricia* species formed a monophyletic group with congeneric species and were clearly distinguished from the sister taxa (Fig. 5). *Henricia epiphysialis* sp. nov. was clearly monophyletic in the *COX1* dataset of 189 bp (Fig. 5A), but the 189 bp *COX1* dataset was too short for species-level identification using DNA barcoding analysis. Therefore, we reanalyzed phylogenetic relationships using a longer *COX1* dataset (537 bp) including 27 species of *Henricia* (Table 1, Fig. 5B). The phylogenetic tree of the 537 bp dataset also revealed that *H. epiphysialis* sp. nov. presented a monophyletic node according to short and long *COX1* datasets analysis (Fig. 5). In addition, each *Henricia* species examined was grouped at the species level. The intra- and inter-specific variations of *Henricia* species from the 537 bp *COX1* dataset were calculated by the Kimura 2-parameter model and presented in Table 3. According to the p-distance result, the average value between *Henricia* spp. and *Echinaster* spp. was 0.208 (Table 3). The range of inter-specific distance in the genus *Henricia* was 0.002–0.188, and the average value of inter-specific distance was 0.110 (Table 4). The inter-specific values of *H. kinkasana* and *Henricia* sp. 3–7 was lower than the relationships of other *Henricia* species (Table 3, 4). The range of *Henricia* species, except for *H. kinkasana* and *Henricia* sp. 3–7, was 0.040–0.188 and the average was 0.114 (Table 4). The range and average of *H. epiphysialis* sp. nov. and other *Henricia* species were 0.056–0.154 and 0.098, respectively (Table 4). Therefore, the molecular analysis results show that *H. epiphysialis* sp. nov. is a new species in the genus *Henricia*.

Discussion

The diagnostic combination of the morphological characteristics of the genus *Henricia* such as spination of the abactinal (primarily on the disk and proximal portion of the arm), adambulacral, and oral plates was found to be consistent and reliable for determining the species (e.g., Hayashi 1940; Djakonov 1950; Clark and Jewett 2010). The shape of the abactinal spines and plate formation exhibited the distinct morphological characteristics of *Henricia epiphysialis* sp. nov. (Figs 3A, C, D, 4A–C). *Henricia epiphysialis* sp. nov. was compared with nine related *Henricia* species (Table 2). This new species has very peculiar abactinal spines that form a hooked crown and

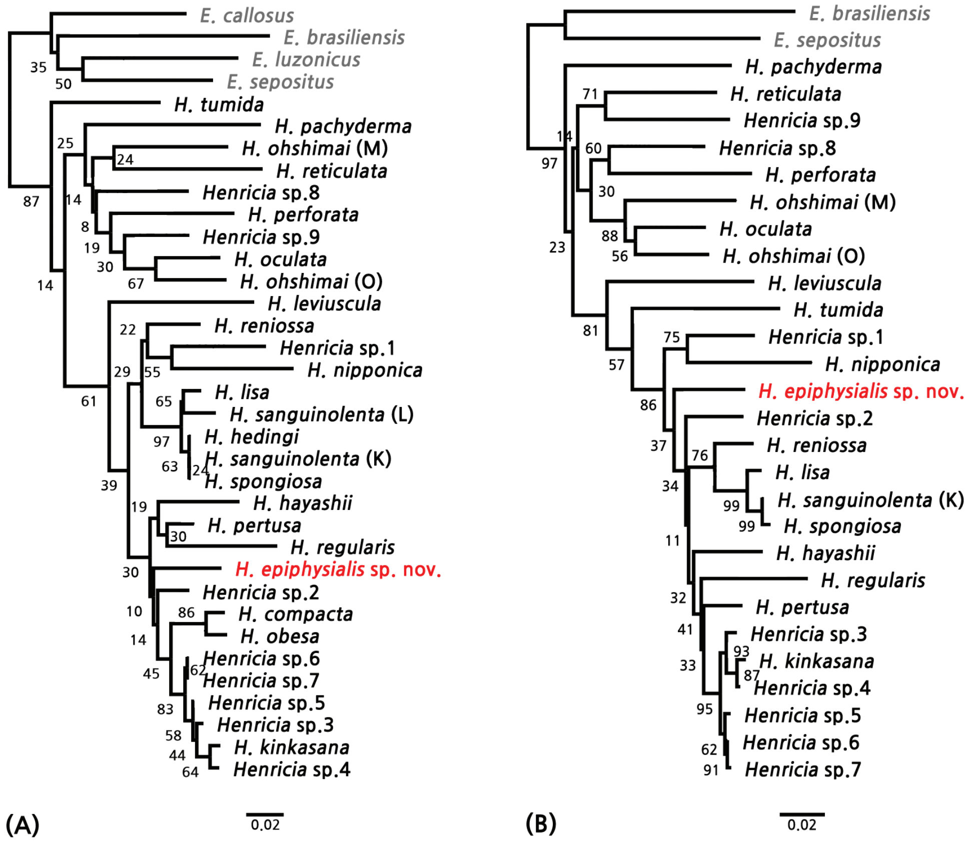

Figure 5. Phylogenetic trees of *Henricia* species including *Henricia epiphysialis* sp. nov. based on Neighbor joining (NJ) **A** NJ tree constructed with 189 bp of *COX1* dataset **B** NJ tree constructed with 537 bp of *COX1* dataset; K, Knott et al., 2018; L, Lopes et al., 2016; M, Misaki, Kanagawa in Wakita et al., 2019; O, Oshoro, Hokakido in Wakita et al., 2019.

can be compared with the robust, coarse abactinal spines of *H. compacta*, *H. leviuscula*, *H. skorikovi*. *Henricia epiphysialis* sp. nov. is superficially similar to *H. leviuscula* in having short and coarse abactinal spines, but differs mainly in the arrangement of the abactinal paxillae (*H. epiphysialis* sp. nov. has less dense abactinal paxillae, whereas *H. leviuscula* has dense abactinal paxillae) and formation of abactinal spines (*H. epiphysialis* sp. nov. has spines forming a hooked crown with small connected apical tips, whereas *H. leviuscula* has spines with solid glassy tips). *Henricia epiphysialis* sp. nov. is morphologically distinguishable from its congeners primarily by the presence of conspicuous epiphyseal ossicles in almost every plate, and also by the distinctive arrangement of the epiphyseal ossicles of the ventrolateral plates, forming a knob-like connection to the adambulacral plates (Fig. 3D). The knob-like form of epiphyseal ossicles in the ventrolateral plates is rarely seen in related *Henricia* species having slender arms with imbricated plates. Our new species, *H. epiphysialis* sp. nov.

Table 2. Comparison of the morphological characteristics of *Henricia epiphysialis* sp. nov. with related *Henricia* species. Morphological data derived from the respective original descriptions, the present study, and Xiao et al. (2011).

	***Henricia epiphysialis* sp. nov.**	***H. compacta* (Sladen, 1889)**	***H. densispina* (Sladen, 1878)**	***H. djakonovi* Chichvarkhin, 2017**	***H. exigua* Hayashi, 1940**	***H. kinkasana* Hayashi, 1940**	***H. leviuscula* (Stimpson, 1857)**	***H. regularis* Hayashi, 1940**	***H. reniossa* Hayashi, 1940**	***H. skorikovi* Djakonov, 1950**
R/r	4.9–5.4	4.4–7.5	2.9–5.5	4.7–5.0	4.2–4.5	4.0–4.8	5.0–6.0	3.1–5.0	5.5–8.0	3.6–5.0
Number of spines of abactinal plates	11–40	up to 45	30 or more	20–30	5–13	5–18	40–60	9–20	15–60 or more	up to 16
Shape of abactinal spines	hooked crown	stout	granular	stout, barrel, bullet tip	slender, pointed tip	fine, slender, pointed tip	short, granular, solid glassy tip	slender, pointed tip	very fine, rough tip	short, robust, thorny tip
Number of abactinal papulae	1–3	1–3	1–3, rarely 5	1 or 2	1–3	single	1–3	single	1 or 2	2–6
Shape of abactinal plates	reniform with conspicuous epiphyseal ossicle	crescentic, compact	subtriangular	cross, oval, triangular, irregular	elliptic, quasi-triangular, quasi-quadrate	crescentic	elliptic, reniform, subquadrate	subquadrate	reniform	slender, rod-like
Shape of actinal plates	rounded cross, elongated cross	quadrilobed, squarish	elongated cross	square pillow	rounded cross, elongated cross	rounded cross, elongated cross	elongated cross, small rod-like	rounded cross	elongated cross, quasi-quadrate	elongated cross, rod-like
Number of actinal papulae	single	single	single	single	single	single	single	single	1 or 2	unknown
Number of adambulacral spines	10–14	5 or 6	11–16	10 or 11	13–15	8–12	15–18	9–13	15–25	7–12
Number of furrow spines	single	2 or 3	single	single	double	single	single	single or double	single	single
Pattern of adambulacral spines (near ambulacral furrow + near ventrolateral plate)	1 long, 2 slender, bluntly pointed + 4–14 slightly shorter	2 or 3 prominent + 4–6 slightly shorter	2 or 3 prominent, bluntly pointed + 4–16 slightly shorter	2 or 3 larger + 4–11 slightly shorter	1 long, 2 spatulate + 4–15 shorter	1 long, 2 slender, bluntly pointed + 4–12 slightly shorter	1 long, 3 stout + 5–18 slightly shorter	1 long, 2 slender, bluntly pointed + 4–13 slightly shorter	1 long, 5 slender + 7–25 slightly shorter	1 long, 2 coarse + 4–12 slightly shorter
Distribution	Korea (East Sea)	southern Australia	Bohai Sea, Yellow Sea, Korea Strait, Tatar Strait, Kurile Island, Japan, Philippines	Rudnaya Bay, Kievka Bay	southern Japan, East China Sea	Japan (off Kinkasan)	Korea (East Sea), Alaska (Kadiak)	East China Sea, Korea (East Sea, Korea Strait, Jeju Island), Japan (Goto Island, Uraga Channel)	Korea (East Sea), Japan (Yezo Strait)	White Sea, Barents Sea, Chesha Bay

Table 3. Pairwise genetic comparison for 537 bp of the mitochondrial *COX1* gene in 27 species of *Henricia* including *Henricia epiphysialis* sp. nov.

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28
H. epiphysialis sp. nov.																												
H. hayashii	0.069																											
H. kinkasana	0.065	0.056																										
H. leviuscula	0.141	0.143	0.132																									
H. lisa	0.084	0.069	0.054	0.114																								
H. nipponica	0.096	0.114	0.112	0.157	0.096																							
H. oculata	0.131	0.143	0.134	0.150	0.139	0.161																						
H. ohshimai 1	0.129	0.143	0.141	0.149	0.136	0.150	0.064																					
H. ohshimai 2	0.142	0.166	0.151	0.161	0.159	0.170	0.079	0.094																				
H. pachyderma	0.154	0.161	0.159	0.160	0.154	0.188	0.136	0.145	0.133																			
H. perforata	0.152	0.164	0.152	0.154	0.167	0.165	0.117	0.141	0.138	0.143																		
H. pertusa	0.067	0.048	0.036	0.136	0.061	0.105	0.141	0.138	0.151	0.173	0.166																	
H. regularis	0.090	0.083	0.071	0.145	0.085	0.118	0.152	0.138	0.165	0.188	0.191	0.065																
H. reniossa	0.077	0.058	0.056	0.114	0.042	0.096	0.136	0.134	0.159	0.154	0.169	0.052	0.083															
H. reticulata	0.135	0.133	0.133	0.171	0.152	0.173	0.120	0.122	0.113	0.131	0.159	0.133	0.146	0.147														
H. sanguinolenta	0.084	0.067	0.056	0.118	0.013	0.092	0.139	0.134	0.159	0.154	0.164	0.063	0.090	0.034	0.150													
H. spongiosa	0.088	0.071	0.061	0.118	0.017	0.092	0.144	0.139	0.164	0.159	0.169	0.067	0.094	0.038	0.154	0.004												
H. tumida	0.104	0.117	0.115	0.161	0.124	0.135	0.137	0.137	0.146	0.156	0.165	0.120	0.133	0.124	0.128	0.124	0.129											
Henricia sp. 1	0.090	0.087	0.077	0.141	0.071	0.086	0.150	0.145	0.162	0.150	0.156	0.081	0.111	0.058	0.160	0.067	0.071	0.133										
Henricia sp. 2	0.062	0.062	0.044	0.120	0.059	0.105	0.141	0.145	0.158	0.159	0.154	0.050	0.081	0.056	0.147	0.056	0.061	0.113	0.077									
Henricia sp. 3	0.058	0.052	0.015	0.123	0.052	0.105	0.136	0.136	0.156	0.156	0.159	0.032	0.069	0.048	0.140	0.055	0.059	0.122	0.073	0.040								
Henricia sp. 4	0.063	0.054	0.006	0.129	0.052	0.110	0.131	0.138	0.149	0.156	0.154	0.034	0.069	0.054	0.137	0.055	0.059	0.113	0.075	0.042	0.009							
Henricia sp. 5	0.058	0.048	0.015	0.127	0.050	0.103	0.141	0.138	0.154	0.152	0.164	0.029	0.058	0.048	0.138	0.053	0.057	0.120	0.073	0.044	0.011	0.013						
Henricia sp. 6	0.056	0.048	0.015	0.123	0.046	0.099	0.136	0.134	0.149	0.149	0.164	0.029	0.058	0.044	0.140	0.048	0.053	0.117	0.069	0.040	0.011	0.013	0.004					
Henricia sp. 7	0.058	0.050	0.017	0.125	0.048	0.096	0.134	0.131	0.147	0.147	0.161	0.031	0.061	0.046	0.138	0.050	0.055	0.120	0.067	0.042	0.013	0.015	0.006	0.002				
Henricia sp. 8	0.141	0.145	0.136	0.131	0.141	0.147	0.096	0.107	0.111	0.145	0.107	0.138	0.147	0.138	0.143	0.134	0.138	0.147	0.138	0.141	0.141	0.134	0.136	0.132	0.129			
Henricia sp. 9	0.145	0.143	0.145	0.148	0.157	0.176	0.107	0.127	0.139	0.147	0.129	0.154	0.166	0.150	0.105	0.152	0.157	0.146	0.168	0.145	0.138	0.138	0.148	0.148	0.150	0.125		
E. brasiliensis	0.220	0.212	0.228	0.210	0.217	0.207	0.227	0.217	0.202	0.190	0.200	0.230	0.230	0.233	0.202	0.220	0.225	0.197	0.217	0.227	0.225	0.230	0.225	0.223	0.220	0.205	0.217	
E. sepositus	0.192	0.190	0.207	0.190	0.207	0.190	0.192	0.195	0.183	0.214	0.192	0.207	0.215	0.210	0.219	0.205	0.205	0.204	0.209	0.190	0.210	0.205	0.210	0.205	0.205	0.178	0.212	0.190

Table 4. The range and average p-distance values of *Henricia* species examined in this study.

Group	Range	Average
H. epiphysialis–other *Henricia* sp.	0.056–0.154	0.098
All of *Henricia* species	0.002–0.188	0.110
All *Henricia* species except for *H. kinkasana* and *Henricia* sp.3–sp.7	0.040–0.188	0.114

is morphologically closer to *H. reniossa*: they share similar reniform abactinal plates, elongated cross shaped actinal plates, but *H. epiphysialis* sp. nov. possessed well-developed epiphyseal ossicles in both abactinal and actinal plates. The molecular analysis supports the morphological similarity by showing both species in the same clade (Fig. 5B).

Other morphological characteristics of *H. epiphysialis* sp. nov., such as the ratio of arm to disk and number of adambulacral spines, are similar to those of *H. kinkasana* which is a slender-rayed species; however, this new species differs mainly in the number of abactinal spines and shape of both abactinal and lateral spines. *Henricia epiphysialis* sp. nov. has 11–40 robust abactinal spines on the abactinal plate, whereas *H. kinkasana* has five to 18 fine, delicate abactinal spines. Moreover, the conspicuous epiphyseal ossicles at the ends of the abactinal and lateral plates are exclusively present in *H. epiphysialis* sp. nov. Therefore, the extension of ossicles in the plate and hooked crown shape of the spines are diagnostic characteristics for this new species.

In this study, we identify a new *Henricia* species based on its morphological characteristics and DNA barcoding. *Henricia epiphysialis* sp. nov. has distinct morphological features and was classified as a new species after comparison with related species. Moreover, the molecular analysis showed that *H. epiphysialis* sp. nov. clearly formed a monophyletic node in a large clade of the genus *Henricia* species (Fig. 5), and the minimum value for the inter-specific distance was significantly higher than the inter-specific distance reported in a previous asteroid DNA barcoding study (Table 3) (Ward et al. 2008). Therefore, the molecular analysis clearly supported the diagnostic morphological identification of *H. epiphysialis* sp. nov. as a new species under the genus *Henricia*. The mitochondrial *COX1* gene is especially useful and effective for the DNA barcoding analyses of *Henricia* species.

Acknowledgements

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RESEARCH ARTICLE

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Revision of the genus *Laophontodes* T. Scott (Copepoda, Harpacticoida, Ancorabolidae), including the description of a new species and a key to species

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Abstract

The description of *Laophontodes volkerlehmanskii* **sp. nov.** (Copepoda, Harpacticoida, Laophontodinae Lang) from the deep sea of the Kairei Field, western Indian Ocean, prompted the examination of the phylogenetic status of *Laophontodes* T. Scott and the relationships within the genus. The allocation of *L. volkerlehmanskii* **sp. nov.** to *Laophontodes* based on diagnostic characters was relatively straightforward, yet phylogenetic analysis of the genus considering 39 morphological characters detected not a single autapomorphy. This indicates that *Laophontodes*, which seems to form a monophylum with *Ancorabolina* George and *Bicorniphontodes* George, Glatzel & Schröder, actually represents the stem-lineage, retaining the characters of the common ancestor without having developed unique derived morphological characters. Most of the 13 known species of *Laophontodes* can be characterised by distinct apomorphies. However, phylogenetic comparison highlights some uncertainties due to the apparent heterogeneous distribution of some derived characters across the species, the weakness of other features, and the fragmentary and inadequate description of several species, which, in combination with the unavailability of type material, prevents a detailed comparison of several phylogenetically relevant characters. Thus, the analysis presented here provides a further step towards understanding the systematic relationships of and within *Laophontodes*, rather than a conclusive answer. Nonetheless, a detailed character discussion and a key to species are given.

Keywords

Crustacea, Indian Ocean, Kairei Hydrothermal Vent Field, Laophontodinae, meiofauna, phylogeny, taxonomy

Introduction

Recent extensive revisions of the Ancorabolidae Sars, 1909 (e.g., George 2006; Gheerardyn and George 2010; Gheerardyn and Lee 2012; George and Müller 2013; George and Gheerardyn 2015; George 2017, 2018; George et al. 2019; Lee and Huys 2019; George 2020), revealed problems in the phylogenetic characterisation of the genus *Laophontodes* Sars, 1894 as a monophylum (cf. George and Gheerardyn 2015; George 2018, 2020; George et al. 2019); despite 13 species being assigned to *Laophontodes* (cf. Lee and Huys 2019; George 2020) not a single autapomorphy has been detected. Although genetic approaches can provide valuable information, molecular data for *Laophontodes* are not available, and morphological analyses are the most robust method for determining phylogenetic relationships. Such an approach must include the detailed descriptions of all species, including new ones, in order to enable the detection of not only species characteristics, but also derived features exclusively shared by all *Laophontodes* species (= synapomorphies), and thus facilitating phylogenetic comparisons (cf. George 2017). In that context, we describe a new *Laophontodes* species, *L. volkerlehmanskii* sp. nov. from the western Indian Ocean. It represents the first member of the genus in that geographic area, and one of few species inhabiting bathyal depths > 2000 m. Apart from a discussion on the systematics within *Laophontodes*, a key to the *Laophontodes* species is given.

Material and methods

Samples were collected during the INDEX 2012 expedition of RV FUGRO GAUSS in December 2012 at the Kairei Field, an active hydrothermal vent field within the Central Indian Ridge (Kihara and Schröter 2013) (Fig. 1). Sampling was undertaken using a TV grab at a depth of 2467 m. The here described *Laophontodes volkerlehmanskii* sp. nov. was collected on 1 December, 2012 at station #I12_36T. The material collected was sieved with a 300 µm mesh and preserved in a solution of 4% buffered formalin, 96% ethanol, and DESS. Centrifugation with 40% Levasil and kaolin was performed three times at 5000 rpm for five minutes to extract the fauna. The individuals gathered were sorted by hand using a Leica M125 stereomicroscope.

Specimens to be studied were embedded in glycerol and put on slides for further investigation. Species identification and drawings were made with the use of a camera lucida on a Leica DMR compound microscope equipped with differential interference contrast optics.

Confocal Laser Scanning Microscopy (CLSM) was used to examine three individuals, two females and one male. The individuals were stained overnight with a

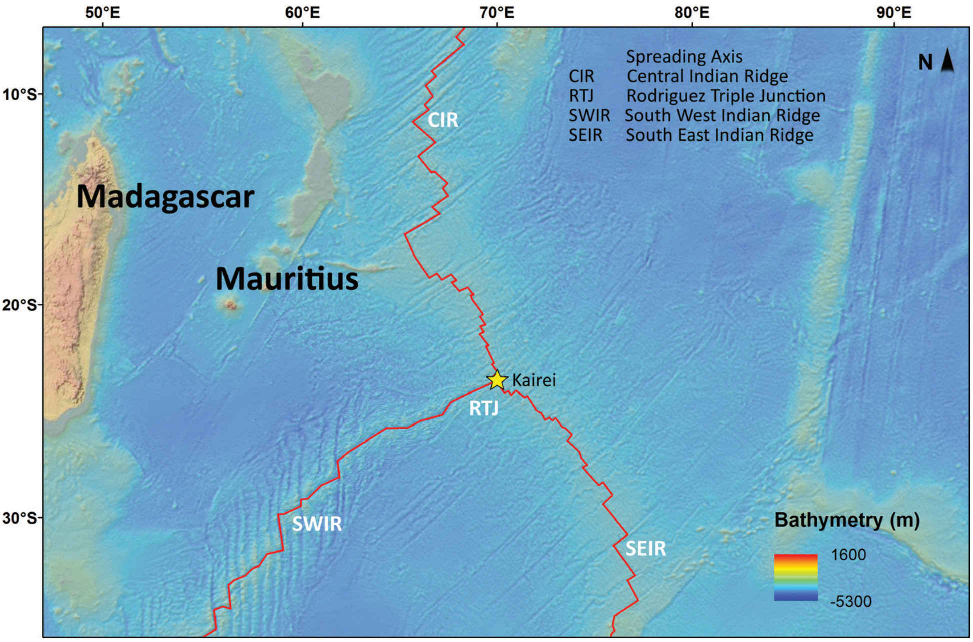

Figure 1. Map showing the place of discovery of the presented species. The star indicates the sampling station at the Kairei ridge, adapted from Dr Klaas Gerdes (Hamburg, Germany).

1:1 solution of Congo Red and Acid Fuchsin adapted from Michels and Büntzow (2010). Specimens were individually mounted in a drop of glycerine surrounded by a transparent, self-adhesive reinforcement ring to prevent direct contact between the specimen and coverslip and, therefore, damage to or distortion of the specimen. Images were generated using a Leica TCS SP5 consisting of a Leica DM5000 B upright microscope and three visible-light lasers. The software used was LAS AF 2.2.1 (Leica Application Suite Advanced Fluorescence). Images were taken with objective HCX PL APO CS 10.0× 0.40 DRY UV at an extinction wavelength of 561 nm with 80% acousto-optic tuneable filter. Using overlapping optical sections, passing through the whole specimen with an ideal number of sections determined by the software, a series of stacked images was generated. Table 1 lists the applied settings. To obtain a three-dimensional representation from selected body parts, the data produced during the CLSM scanning was processed with the software Drishti (http://anusf.anu.edu.au/Vizlab/drishti/). The obtained images were finalised with maximum projection and Adobe Photoshop CS6 for adjusting colour, contrast and brightness. The type material is kept in the collection of the Senckenberg Forschungsinstitut und Naturmuseum, Frankfurt am Main (Germany).

The phylogenetic analysis strictly follows Hennig (1982) and Ax (1984, 1988, 1995) as explained by George (2020) and without the application of any computer-based cladistic programs. Consequently, Fig. 11 is not a computer-generated cladogram; instead, it is a manually generated clear presentation of the results of the phylogenetic discussion.

Table 1. Confocal laser scanning microscopy (CLSM) settings. Ch1 = detection channel 1.

Acquisition resolution	2048 × 2048
Numerical aperture	0.4
Excitation beam splitter	DD 488/561
Detected emission wavelength (nm)	Ch1: 570–629 Ch2: 629–717
Detector gain	544 and 509 V
Amplitude offset	-1.7 and 0.8%
Pinhole aperture (µm)	53.0

General terminology follows Lang (1948), Huys and Boxshall (1991), and Huys et al. (1996). Terminology referring to phylogenetic aspects follows Ax (1984); the terms "telson" and "furca" are adopted from Schminke (1976).

Abbreviations used in the text:

A1:	antennule;	**md:**	mandible;
A2:	antenna;	**mx:**	maxilla;
aes:	aesthetasc;	**mxl:**	maxillule;
cphth:	cephalothorax;	**mxp:**	maxilliped;
enp-1–enp-3:	endopodal segments 1–3;	**n:**	number of specimens;
exp-1–exp-3:	exopodal segments 1–3;	**P1–P6:**	swimming legs 1–6;
FR:	furcal rami;	**R:**	rostrum;
GDS:	genital double somite;	**STE:**	subapical tubular extension.
GF:	genital field;		

Results

Subclass Copepoda Milne Edwards, 1840
Order Harpacticoida Sars, 1903
Family Ancorabolidae Sars, 1909
Subfamily Laophontodinae Lang, 1944

Genus *Laophontodes* T. Scott, 1894

Species composition. *L. typicus* T. Scott, 1894 (type species); *L. antarcticus* Brady, 1918, *L. georgei* Lee & Huys, 2019, *L. gertraudae* George, 2018, *L. macclintocki* Schizas & Shirley, 1994, *L. monsmaris* George, 2018, *L. mourois* Arroyo, George, Benito & Maldonado, 2003, *L. sabinegeorgeae* George & Gheerardyn, 2015, *L. sarsi* George, 2018, *L. scottorum* George, 2018, *L. spongiosus* Schizas & Shirley, 1994, *L. whitsoni* T. Scott, 1912 (cf. George 2020); *species inquirenda*: *L. propinquus* Brady, 1910.

Remarks. Lee and Huys (2019) listed 18 species in *Laophontodes* – 15 valid species plus one *species incertae sedis* (*L. propinquus* Brady, 1910) and two *species inquirendae* (*L. antarcticus* Brady, 1918, *L. ornatus* Krishnaswamy, 1957). The number of species was updated by George et al. (2019), who established the genus *Bicorniphontodes*

George, Glatzel & Schröder, 2019 to include the then newly described *B. clarae* George, Glatzel & Schröder, 2019, along with *Laophontodes bicornis* A. Scott, 1896, *L. hamatus* (Thomson, 1883), *L. horstgeorgei* George & Gheerardyn, 2015, and *L. ornatus* Krishnaswamy, 1957. George et al. (2019) reduced the number of species allocated to *Laophontodes* to 14. Of these, *Laophontodes brevis* Nicholls, 1944 was excluded from the current analysis: although Lee and Huys (2019: 367) are certainly right when insisting on its validity as a species, since Lang (1965) did not formally synonymise *L. brevis* with *L. bicornis* (now *Bicorniphontodes bicornis*), the remarkable similarity of *L. brevis* with *B. bicornis* noted by Lang (1965) clearly points to its affiliation to *Bicorniphontodes* instead to *Laophontodes*. Therefore, we follow the list of 13 species of *Laophontodes* as provided by George (2020).

***Laophontodes volkerlehmanskii* sp. nov.**

http://zoobank.org/34839233-C919-4892-9CA9-8FAFEFCF0D70
Figs 2–10

Locus typicus. Indian Ocean, Central Indian Ridge, Kairei Field, station #I12_36T, geographic position 25°19.240'S, 70°02.433'E, 2467 m depth.

Type material. Four females and four males collected during research cruise INDEX 2012 on December 1st, 2012. Holotype: female, not dissected, on one slide, collection number SMF 37216/1; paratype 1 (allotype): male, not dissected, on one slide, collection number SMF 37217/1; paratype 2: female, dissected and mounted onto 15 slides, collection number SMF 37218/1–15; paratype 3: male, dissected and mounted onto two slides, collection number SMF 37219/1–2; paratype 4: male, not dissected, on one slide, collection number SMF 37220/1; paratype 5: male, not dissected, on one slide, collection number SMF 37221/1; paratype 6: female, not dissected, on one slide, collection number SMF 37222/1; paratype 7: female, not dissected, on one slide, collection number SMF 37223/1.

Description. Female: Habitus (Figs 2A, 3A, B) cylindrical, body length (R to end of FR) (median value) = 399 µm (390–405 µm; *N* = 3). R small, fused to cphth, with 2 sensilla (one sensillum missing in Fig. 2A) and 1 apical tube pore. Cphth reaching more than 25% of total body length, with posterior swelling on each side; dorsally covered by sensilla, those on posterior margin arising from socles. Body somites clearly distinct. Last thoracic and first abdominal somites fused forming the GDS, juncture seen as dorsal serration. Posterior margins of free body somites, excluding telson, serrated dorsally, and with sensilla arising from small socles; P2–P5-bearing somites additionally with 1 dorsal tube pore centrally; P6 and P7-bearing somites carrying 2 dorsal tube pores centrally. Telson (Figs 2A, 3A, B, 4A) slightly smaller than preceding somite, with FR set widely apart. Anal operculum (Figs 4A, 5A) with distinct, strong apical spinules; basally with pair of sensilla and additional spines above bases of FR.

FR (Figs 4A, 5A) slender, about 4 times as long as wide, with distal tube pore and 7 bare setae: seta II dorsal to, and twice as long as I; III subapical; IV and V apical and fused at base, V very long and narrow, distal 1/3 bipinnate; VI apical on inner margin, bare and short; VII dorsal, tri-articulated.

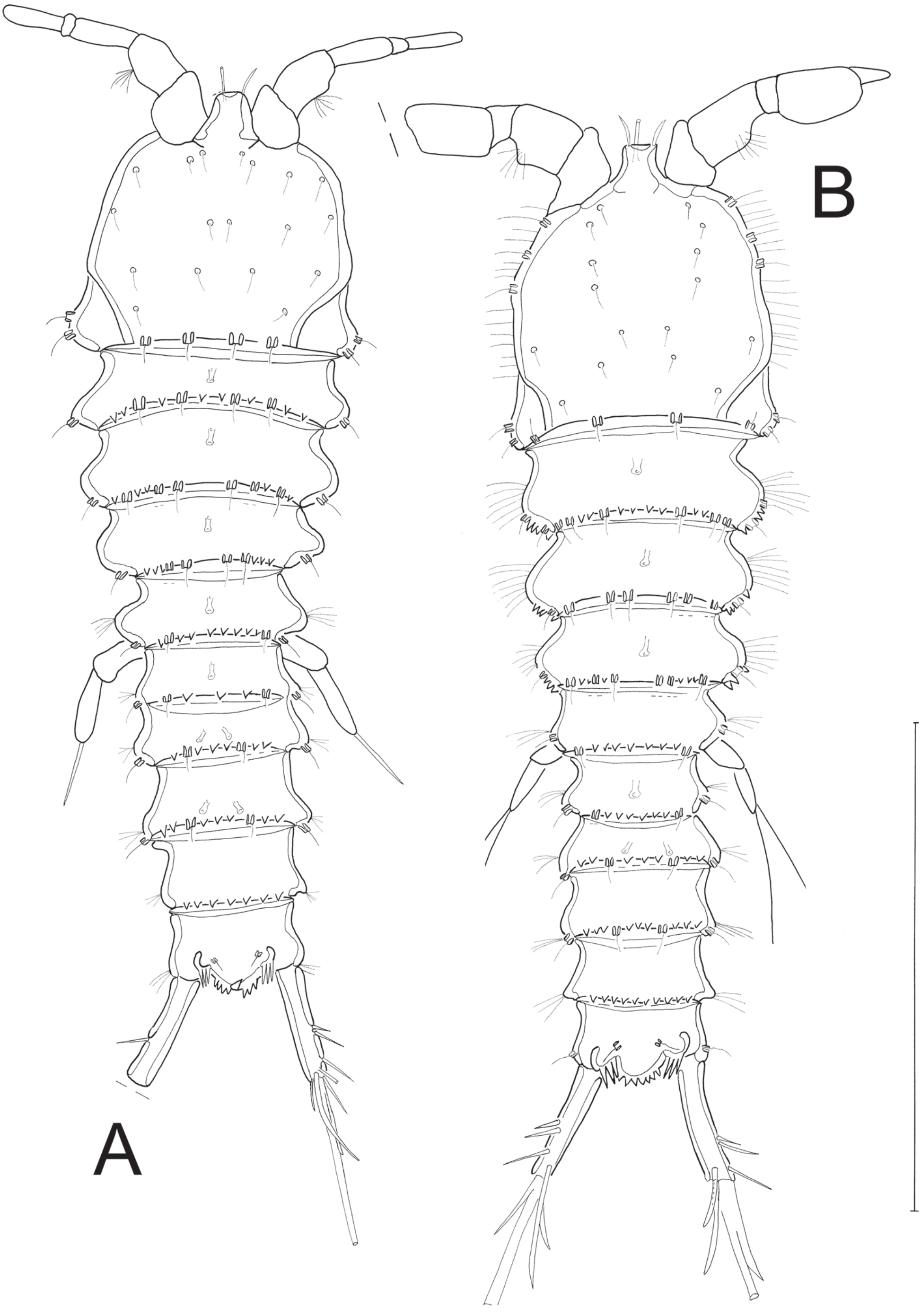

Figure 2. *Laophontodes volkerlehmanskii* sp. nov. **A** female holotype (SMF 37216/1), habitus, dorsal view **B** male paratype 1 (allotype) (SMF 37217/1), habitus, dorsal view. Scale bar 200 µm.

Figure 3. *Laophontodes volkerlehmanskii* sp. nov. Confocal laser scanning microscopy images of **A** female paratype 6 (SMF 37222/1), habitus, dorsal view **B** female paratype 7 (SMF 37223/1), habitus, ventral view. Scale bar: 25 µm.

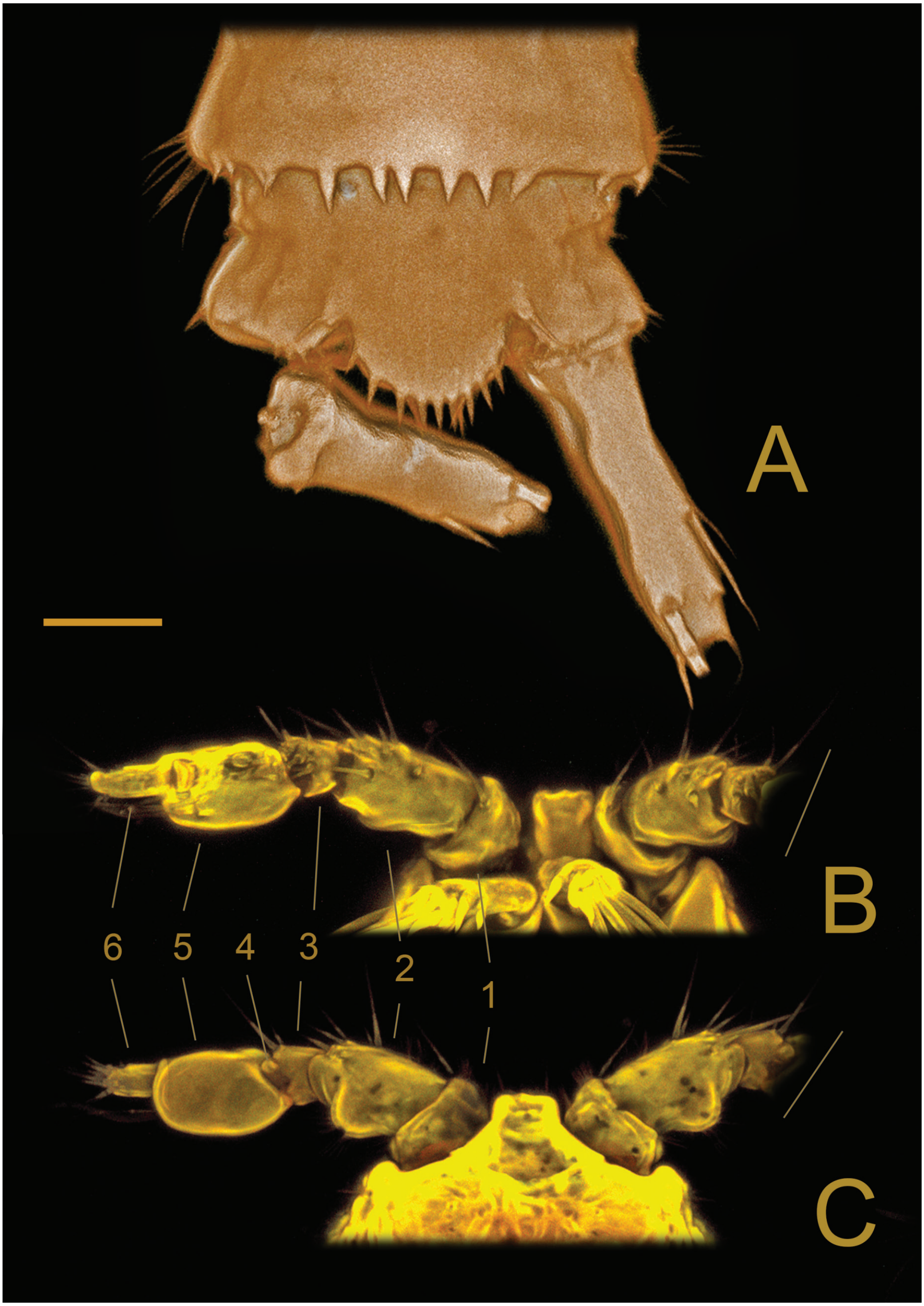

Figure 4. *Laophontodes volkerlehmanskii* sp. nov. Three-dimensional representation (Drishti software) based on confocal laser scanning microscopy images of Female paratype 6 (SMF 37222/1) **A** anal operculum, dorsal view; Confocal laser scanning microscopy images of male paratype 4 (SMF 37220/1) **B** A1, ventral view **C** male paratype 4 (SMF 37220/1), A1, dorsal view, numbers refer to antennular segments Scale bar: 400 µm (**A**); 25 µm (**B, C**).

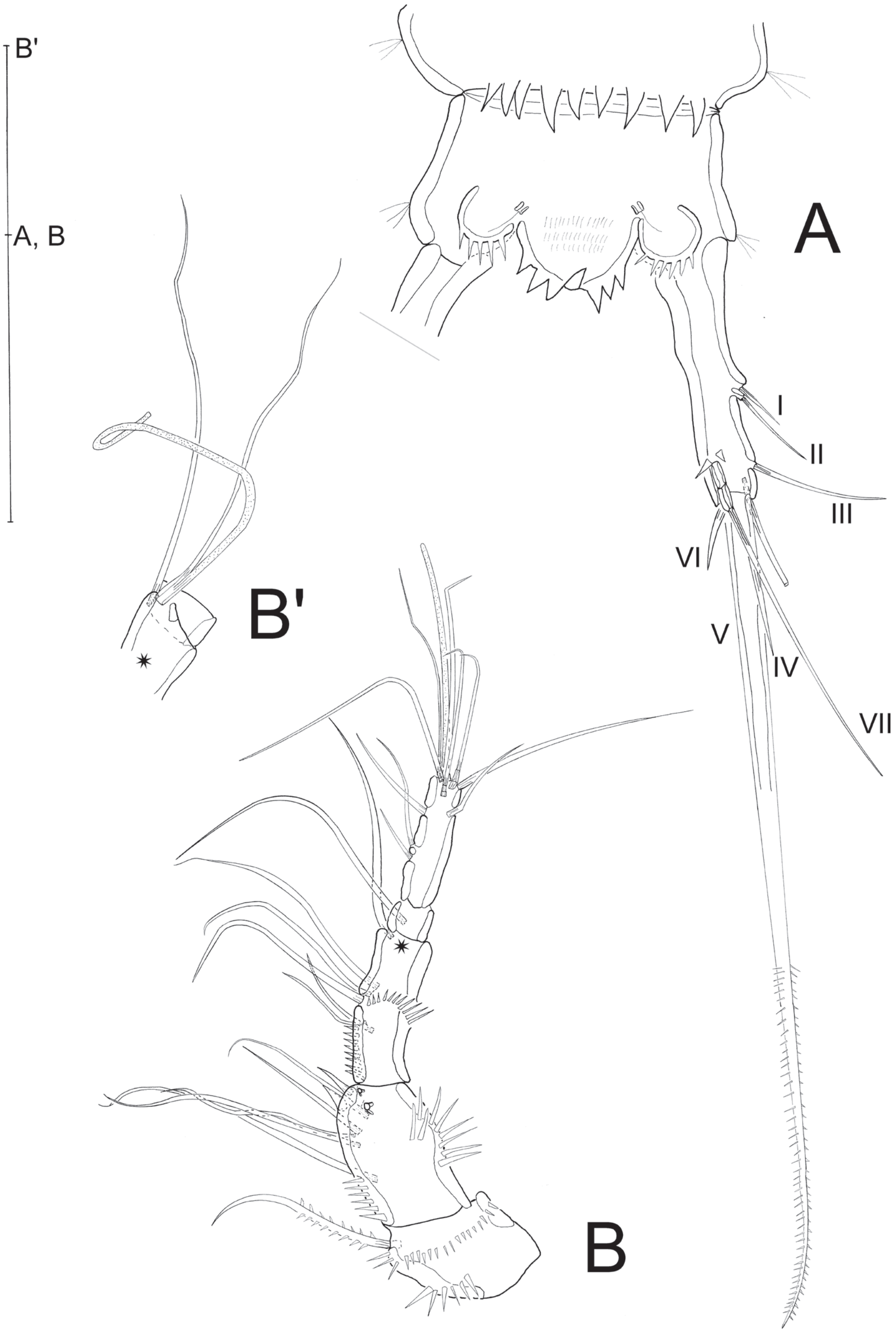

Figure 5. *Laophontodes volkerlehmanskii* sp. nov., female holotype (SMF 37216/1) **A** telson and right furcal ramus, dorsal view; Roman numbers indicate furcal setae, **B** A1, **B'** Posterior margin of fourth antennular segment, showing projection that bears the acrothek. Scale bar: 50 µm.

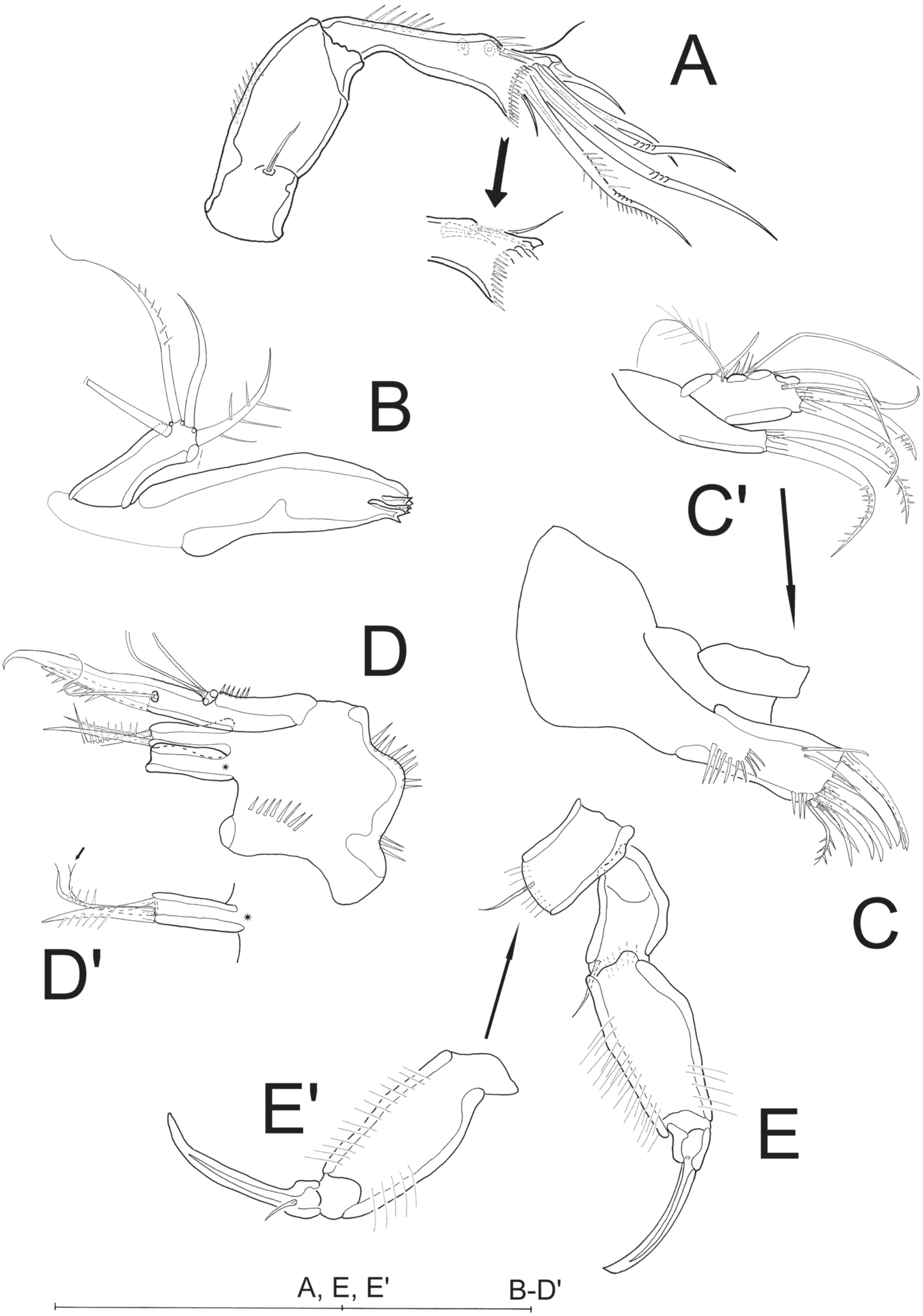

Figure 6. *Laophontodes volkerlehmanskii* sp. nov., female paratype 2 (SMF 37218/1–15) **A** A2 **B** Md **C** Mxl gnathobase **C'** Mxl coxa and basis **D** Mx **D'** Mx, proximal endite; arrow pointing to cleft tip **E** Mxp **E'** Mxp (counterpart), showing minute accompanying seta. Scale bar: 50 µm.

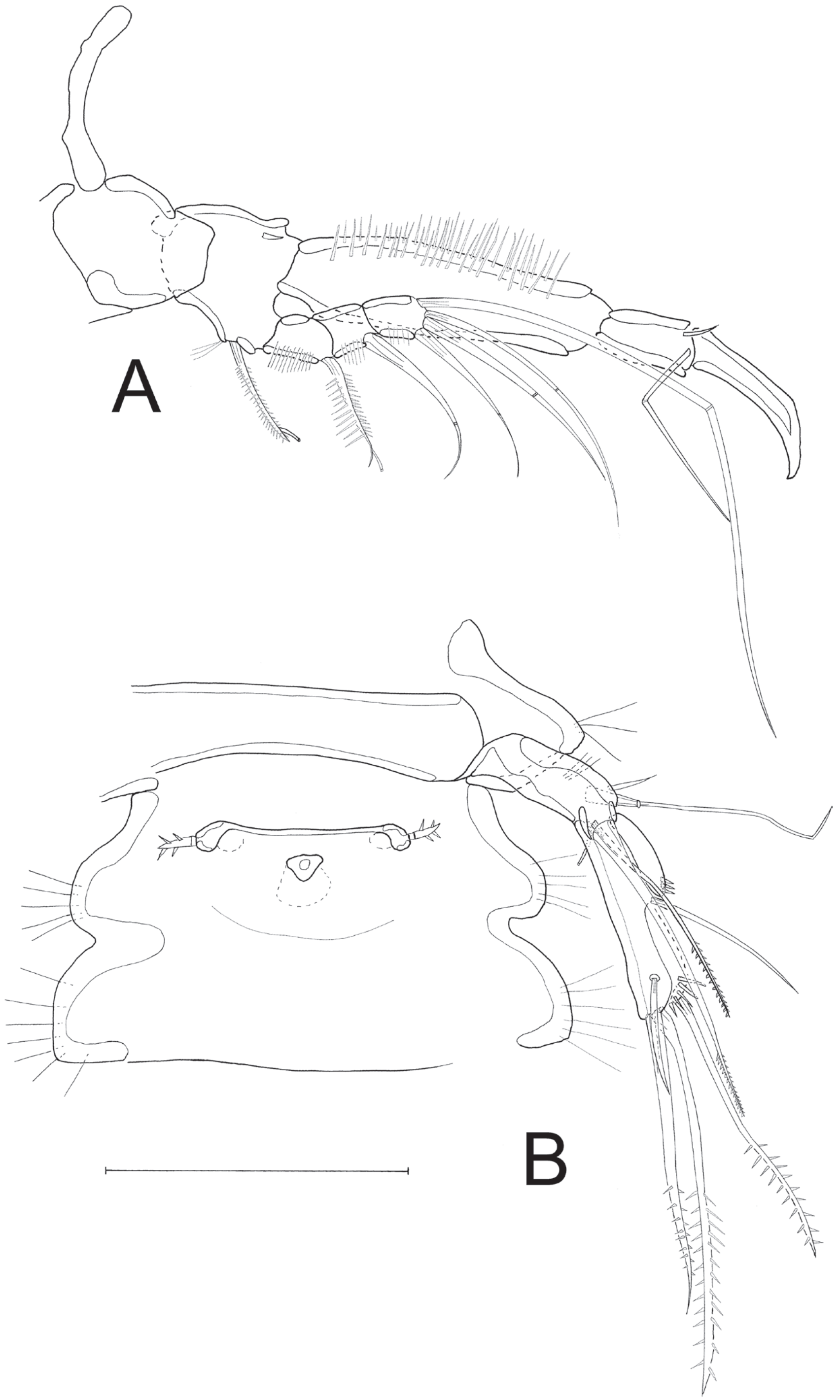

Figure 7. *Laophontodes volkerlehmanskii* sp. nov., female paratype 2 (SMF 37218/1–15) **A** P1 **B** P5 and GF with minute P6. Scale bar: 50 µm.

A1 (Fig. 5B, B') 5-segmented. First segment carrying 1 bipinnate seta and 2 rows of spinules on apical edge below seta; second segment with 9 setae (2 setae broken in Fig. 5B) and 1 row of spinules each on outer and inner margin; third segment with 7 bare setae and 1 aes (fused to 1 seta) (Fig. 5B'); fourth segment partly overlapped by preceding one (Fig. 5B'), with 1 bare seta; fifth segment with 10 bare setae, 2 of which forming an apical trithek with1 aes. Setal formula: 1-1/2-9/3-6+(1+aes)/4-1/5-8+(2+aes).

A2 (Fig. 6A). Allobasis without abexopodal seta. Exopod represented by minute bare seta. Endopod with 2 rows of inner spinules – 1 subapical and 1 proximal – ; additionally, with 2 bare spines and 1 fine bare seta accompanied by 4 anterior spinules; apically with 5 setae, of which 3 geniculated and 1 biplumose.

Md (Fig. 6B) with slender gnathobase bearing 4 teeth; palp 1-segmented, with 5 setae (1 missing, and 1 damaged in Fig. 6B), of which 1 biplumose and 1 unipinnate.

Mxl (Fig. 6C, C'). Praecoxal arthrite (Fig. 6C) with 1 row of spinules basally, 8 bare apical spines and 1 subapical biplumose seta, additionally with 2 surface setae; coxa (Fig. 6C') with 2 bipinnate apical setae; basis, endopod and exopod fused to single lobe (Fig. 6C') carrying 2 unipinnate and 1 bare apical seta, 1 bare subapical seta, 3 bare and 1 bipinnate seta and few outer spinules.

Mx (Fig. 6D, D'). Syncoxa bearing 3 rows of spinules and 2 endites. Proximal endite with 2 plumose setae and 1 bare seta with cleft tip (arrow in Fig. 6D'), distal endite with 1 plumose and 2 bare setae. Allobasis distinct, terminally with strong claw accompanied by 1 plumose and 1 fine, bare seta. Endopod 1-segmented, knob-like, with 2 bare setae.

Mxp (Fig. 6E, E') prehensile; syncoxa bearing 1 bare seta and single row of spinules apically (Fig. 6E); basis with 1 row of spinules on inner and outer margins; endopod drawn out into strong claw basally accompanied by minute, bare seta (Fig. 6E').

P1 (Figs 3B, 7A) with slender and bow-like intercoxal sclerite, and large, triangular praecoxa (Fig. 3B); coxa and basis with slight longitudinal elongation, outer margin of basis forming pedestal for exopod, with 1 biplumose outer seta carrying STE, and 1 minute anterior inner seta. Endopod 2-segmented, enp-1 strong and elongate, with 2 rows of spinules on inner margin; enp-2 small, approximately 1/3 the length of enp-1, apically with 1 strong claw, 1 long, slender, geniculated seta, and 1 minute seta. Exopod 3-segmented, less than half the length of endopod, each segment with outer spinules, exp-1 carrying 1 biplumose outer seta with STE; exp-2 with 1 bare geniculated outer seta; exp-3 apically with 4 bare, geniculated setae.

P2–P4 (Fig. 8A–C) with transversely elongated bases with outer margin bearing 1 long seta, the latter bipinnate in P2, bare in P3 and P4; exopods 3-segmented, endopods 2-segmented. All exopodal segments with outer row of robust spinules, and fine inner spinules. Exp-1 and exp-2 with 1 bipinnate outer spine; exp-3 with 3 pinnate outer spines, apically with 1 spine, whose inner margin plumose and outer margin pinnate, and 1 slender biplumose seta. Endopods narrow, enp-1 small, without spinules or setae; enp-2 elongate, of P2 and P4 with spinules, P2–P4 with 2 apical setae, both biplumose in P3 and P4, inner apical seta bare in P2. See Table 2 for setal formula.

Table 2. *Laophontodes volkerlehmanskii* sp. nov., setation of P2–P4. Roman numerals indicate outer spines.

	Exp-1	Exp-2	Exp-3	Enp-1	Enp-2	Enp-3
P2	I-0	I-0	III-2-0	0	0-2-0	–
P3 female	I-0	I-0	III-2-0	0	0-2-0	–
P3 male	I-0	I-0	III-2-0	0	0 (apophysis)	0-2-0
P4 female	I-0	I-0	III-2-0	0	0-2-0	–
P4 male	I-0	I-0	III-2-0	0	I-2-0	–

P5 (Fig. 7B) with short setophore on slender baseoendopod carrying 1 long bare seta and a few spinules; endopodal lobe reduced, represented by 2 bipinnate setae. Exopod fused to baseoendopod, slender, with 1 bare outer seta, 1 bare seta displaced to anterior surface, and 3 plumose setae – 1 subapical and 2 apical.

GF (Fig. 7B) with single gonopore. P6 strongly reduced, limbs fused into single small plate, with pair of minute bipinnate spines.

Male: The male differs from the female in the following characters: habitus, A1, P3 and P4 endopod, and P5.

Habitus (Figs 2B, 9) as in female, but slightly longer, body length (from R to FR) (median value) = 402 µm (378–426 µm; *N* = 2); cphth with more dorsal sensilla than female; with antero-lateral sensilla arising from socles.

A1 (Figs 4B, C, 10A, A') 6-segmented, chirocer; first segment with 1 bipinnate seta and 3 rows of spinules; second segment with 9 bare setae (one seta missing in Fig. 10A), and a row of short spinules; third segment with 6 bare setae (one seta missing in Fig. 10A), and single row of spinules, segment partially overlapping fourth and fifth segment; fourth segment (Fig. 4C, * in Fig. 10A) minute, almost completely covered by fifth segment, with 1 bare seta; fifth segment (Fig. 10A') swollen, with 9 setae (1 biplumose, 8 bare), 2 of which form an acrothek with 1 aes, cuticle thorn-like at upper margin; sixth segment with 10 bare setae (1 seta missing in Fig. 10A'), 2 of which form an acrothek with 1 small aes. Setal formula: 1-1/2-8/3-6/4-1/5-7+(2+aes)/6-8+(2+aes).

P3 exopod as in female, endopod (Fig. 10B) 3-segmented; enp-1 minute and unarmed; enp-2 longest, with rows of spinules on inner and outer margins, lacking setae but inner apical margin with curved apophysis reaching to 2/3 the length of enp-3; enp-3 about 2/3 the length of enp-2, with 2 biplumose apical setae.

P4 exopod as in female, endopod (Fig. 10C) 2-segmented; enp-2 with 1 additional flexible outer spine accompanied by few spinules at its base; apically with 2 biplumose setae. The setal formula for P3 and P4 is given in Table 2.

P5 (Fig. 10D) baseoendopod longer than broad, with 1 outer seta arising from short setophore; endopodal lobe incorporated into basal part of baseoendopod and represented by 1 long seta with bipinnate distal half; exopod not fused to baseoendopod, with 1 outer unipinnate seta, and 2 bipinnate setae – 1 subapical and bearing STE, and 1 apical.

Etymology. The epithet *volkerlehmanskii* is given in dedication to the 60th birthday of LMA Lehmanski's father Volker Lehmanski (Gelsenkirchen, Germany).

Diagnostic key to the species of *Laophontodes*[*]

1 Body slender, cylindrical; cphth about 1/4th of total body length (incl. FR) **2**

– Body compact, partially compressed dorsoventrally; cphth about 1/3rd of total body length (incl. FR) .... ***Laophontodes scottorum* George, 2018**

2 Second antennular segment with flat posterior surface **3**

– Second antennular segment with posterior surface produced into bump..... **4**

3 Mxp of moderate size; P3 and P4 exp-3 with 1 inner seta; telson not overlapped by preceding somite dorsally; male antennule subchirocer .... ***Laophontodes macclintocki* Schizas & Shirley, 1994**

– Mxp extremely strengthened; P3 and P4 exp-3 with 2 inner setae; telson overlapped by preceding somite; male antennule chirocer .... ***Laophontodes monsmaris* George, 2018**

4 Apical claw on P1 enp-2 narrow and thin; male antennule subchirocer....... **5**

– Apical claw on P1 enp-2 wide and thickened; male antennule chirocer or subchirocer **6**

5 Pedigerous and abdominal somites dorsally with fine ripples; P2–P4 exp-3 with 1:2:2 inner setae...................... ***Laophontodes whitsoni* T. Scott, 1912**

– No ripples on body somites, abdominal somites except telson dorsally with H-like cuticular structures; P2–P4 with 0:0:1 inner setae............................ .... ***Laophontodes spongiosus* Schizas & Shirley, 1994**

6 P2 and P3 exp-2 with 1 inner seta, P3 and P4 exp-3 with 2 inner setae; rostrum with setulose tuft frontally; pedigerous and abdominal somites with fine ripples dorsally; abdominal somites except telson with paired cuticular ridges dorsally .... ***Laophontodes sabinegeorgeae* George, 2018**

– These characters not combined **7**

7 P4 exp-3 inner apical seta short, flagelliform, bare **8**

– P4 exp-3 inner apical seta long, biplumose... **10**

8 P2 and P3 exp-3 inner apical seta short, flagelliform, bare .... ***Laophontodes sarsi* George, 2018**

– P2 and P3 exp-3 inner apical seta long, biplumose..................................... **9**

9 P2–P4 exp-3 without inner setae; P4 endopod 1-segmented; inner margin of male P3 enp-3 straight................ ***Laophontodes georgei* Lee & Huys, 2019**

– P2–P4 exp-3 with 1 inner seta; P4 endopod 2-segmented; inner margin of male P3 enp-3 bulged out.. .... ***Laophontodes mourois* Arroyo, George, Benito & Maldonado, 2003**

10 P1 exp-1 outer spine unipinnate, comb-shaped, with strong outer pinnae; anal operculum with row of fine spinules on apical margin **11**

– P1 exp-1 outer spine bipinnate, of normal shape; anal operculum with few strong spinules on apical margin...... ***Laophontodes volkerlehmanskii* sp. nov.**

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[*] *L. antarcticus* Brady, 1918 and *L. propinquus* Brady, 1910 excluded

11 P2–P4-bearing somites with fine ripples dorsally; maxillipedal claw without accompanying minute seta; female P5 baseoendopodal inner seta fish-bone-like..**Laophontodes typicus T. Scott, 1894**

– P2–P4-bearing somites without ripples; maxillipedal claw with accompanying minute seta; female P5 baseoendopodal inner seta bipinnate, of normal shape ....**Laophontodes gertraudae George, 2018**

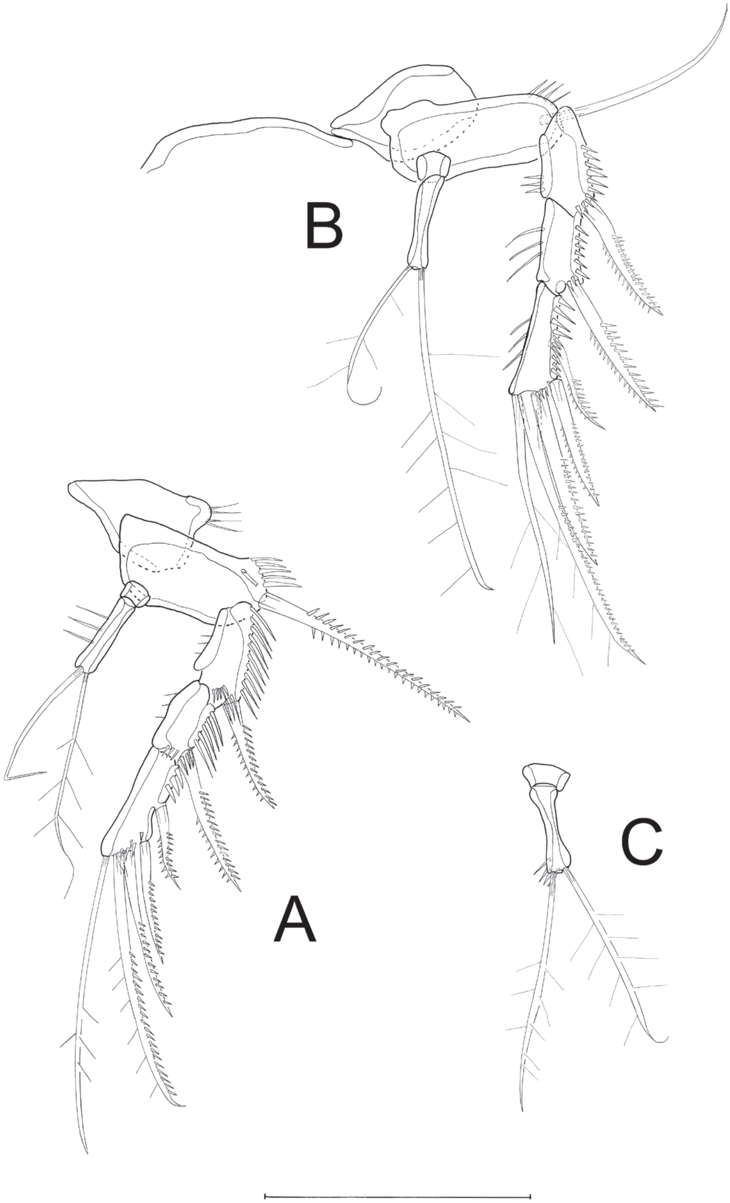

Figure 8. *Laophontodes volkerlehmanskii* sp. nov., female paratype 2 (SMF 37218/1–15) **A** P2 **B** P3 **C** P4 endopod. Scale bar: 50 µm.

Figure 9. *Laophontodes volkerlehmanskii* sp. nov. Confocal laser scanning microscopy images of male paratype 4 (SMF 37220/1) **A** habitus, dorsal view **B** habitus, ventral view. Scale bar: 25 µm.

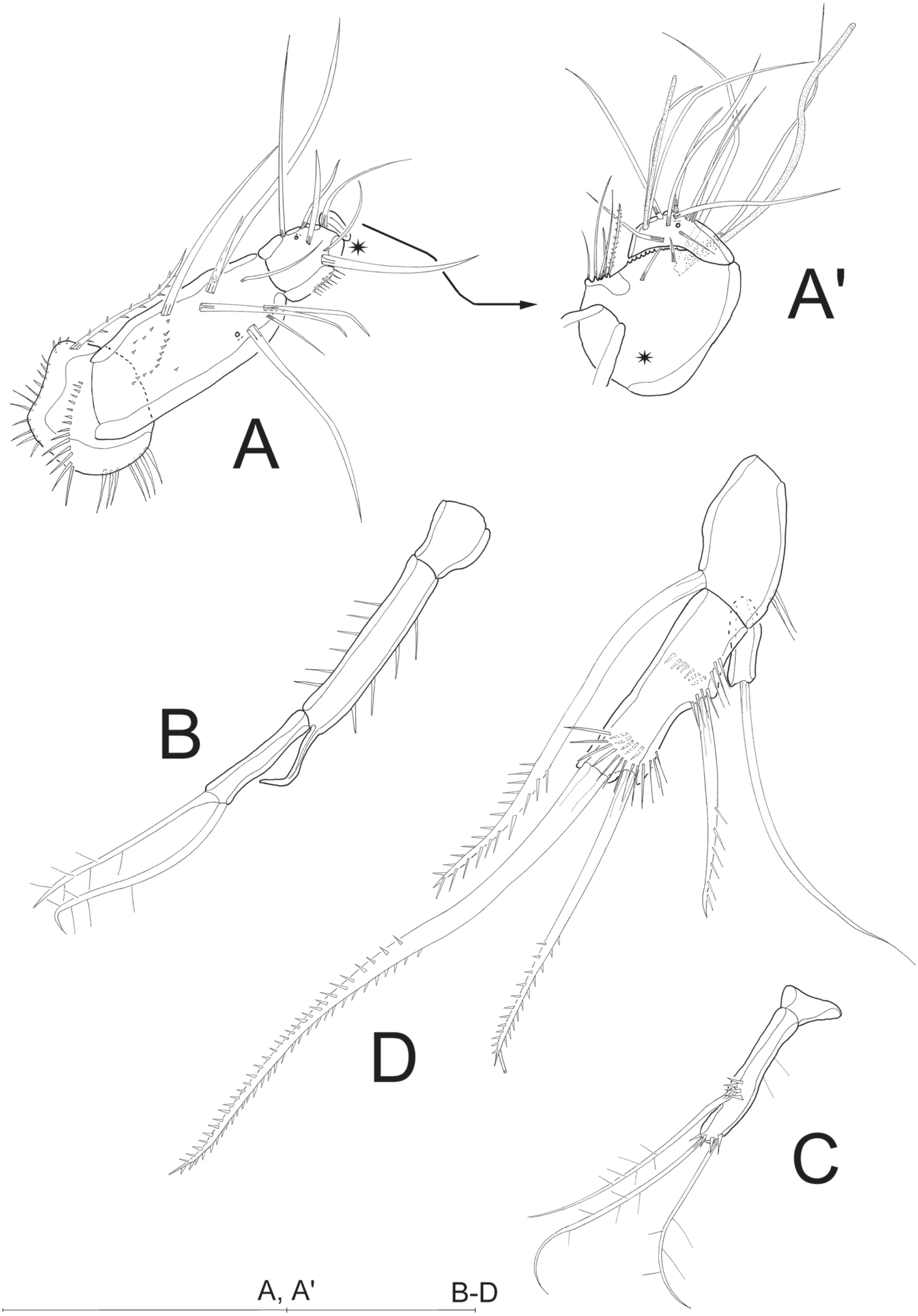

Figure 10. *Laophontodes volkerlehmanskii* sp. nov., male paratype 3 (SMF 37219/1–2) **A** A1 **A'** fifth and sixth antennular segment **B** P3 endopod **C** P4 endopod **D** P5. Scale bar: 50 µm.

Discussion

Laophontodes as "survivor" of the stem-lineage

The type genus of the Laophontodinae – *Laophontodes* – is the only representative of that subfamily which cannot be characterised by autapomorphies (cf. George and Gheerardyn 2015; George 2018, 2020). This causes certain complications, especially as *Laophontodes* comprises 13 species (cf. George 2020), plus the herein described *L. volkerlehmanskii* sp. nov.. As discussed by George (2020), *Laophontodes* seems to be closely related to *Ancorabolina* George, 2006 and *Bicorniphontodes* George, Glatzel & Schröder, 2019. These three genera presumably originate from a common ancestor, which developed one derived, apomorphic, feature (character 1 in Table 3 and below) [supposed ancestral, plesiomorphic, state in square brackets]:

1. A2 exopod lost and represented by 1 tiny seta only [A2 exopod 1-segmented, at least knob-like].

For a detailed discussion of character 1, see George (2020). Since all representatives of the three named genera share that apomorphy, George (2020) interpreted it as autapomorphic for the monophylum [*Ancorabolina – Bicorniphontodes – Laophontodes*] (Fig. 11). *Ancorabolina* and *Bicorniphontodes* share one further apomorphy, viz. the postero-lateral cuticular processes on the cephalothorax and thus form sister-groups, but are otherwise characterised by distinct autapomorphies (George 2020). However, this is not the case for *Laophontodes*. Species can only be assigned to *Laophontodes* using diagnostic characters and this resulted in *Laophontodes* becoming a conglomeration of many, at least partly, unrelated species. Consequently, several authors have excluded species from *Laophontodes*, placing them in newly erected and phylogenetically well-justified distinct genera (e.g., Lang 1965; George 2017: *Paralaophontodes* Lang, 1965; Conroy-Dalton 2004: *Lobopleura* Conroy-Dalton, 2004; Gheerardyn and Lee 2012: *Calypsophontodes* Gheerardyn & Lee, 2012; George et al. 2019: *Bicorniphontodes*; Lee and Huys 2019: *Rostrophontodes* Lee & Huys, 2019, *Lobopleura*). However, as noted by George (2020), it is still not possible to satisfactorily resolve the relationships between those species remaining in *Laophontodes*.

Even with the addition of *L. volkerlehmanskii* sp. nov. as the 14th species, we could not identify a derived feature to support the monophyletic status of *Laophontodes*. The apparent lack of shared morphological novelties within *Laophontodes* suggests that the taxon may represent the stem-lineage, retaining the derived characteristics of the common [*Ancorabolina – Bicorniphontodes – Laophontodes*]-ancestor, having "failed" to develop its own derived characters (Fig. 11). Whilst this is not uncommon (cf. Ax 1984; Sudhaus and Rehfeld 1992), the authors believe this might be the first evidence of a surviving stem-lineage in the Harpacticoida. It remains to be seen if future (molecular genetic) studies may support this hypothesis.

Possible relations within *Laophontodes*

Phylogenetic relationships within *Laophontodes* cannot be resolved unambiguously. This is due to several reasons. For instance, the type material of many species is no longer available, preventing re-examination and comparison of most morphological characters. Moreover, as indicated by several authors (e.g., George and Gheerardyn 2015; George 2017, 2018, 2020; George et al. 2019; Lee and Huys 2019), species descriptions (especially, older publications) are fragmentary and of poor quality, precluding detailed comparisons between species. Nevertheless, such descriptions may be occasionally satisfactorily resolved, as shown below for character 2 (Table 3).

Laophontodes antarcticus and *L. propinquus species inquirenda* were excluded from the phylogenetic analysis presented herein, due to the fragmentary and imprecise descriptions by Brady (1918 and 1910, respectively), and the absence of type material for re-examination.

Careful examination of the remaining 12 species revealed 38 morphological characters as phylogenetically relevant. They are listed in Table 3 (characters 2–39) and are discussed in detail below. Four out of the 38 apomorphies are considered to be convergent (4, 5, 17, 30; underlined in Fig. 11), and the remaining 34 characters as unambiguous. The result of this phylogenetic analysis is graphically summarised in Fig. 11.

A group of nine species within *Laophontodes* (Table 3) share two derived features:

2. Male A1 6-segmented, chirocer [7-segmented, subchirocer];
3. Female P4 endopod with outer seta/spine lost [outer seta/spine still present].

These species (*L. sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., *L. georgei*, *L. gertraudae*, *L. spongiosus*, *L. mourois*, *L. typicus*, *L. monsmaris*; Fig. 11) have lost the penultimate segment of the male A1, which therefore changes from subchirocer to chirocer (character 2) (not yet confirmed for *L. sarsi*, because the males remain unknown). Only three species, namely *Laophontodes macclintocki*, *L. sabinegeorgeae*, and *L. whitsoni*, retain the plesiomorphic 7-segmented, subchirocer male A1. Additionally, the description by Schizas and Shirley (1994) for *L. spongiosus* is contradictory; they state that the male A1 is subchirocer, but describe only one segment after the geniculation, which characterises it as a chirocer A1. Therefore, in Table 3 character 2 is marked with "1?" for *L. spongiosus*.

The derived chirocer condition is hypothesised as synapomorphic for the nine species and this is supported by the concurrent appearance of character 3, viz. the loss of the outer element of the female P4 endopod. Although the reduction of setae/spines occurs frequently and often independently in Harpacticoida, their simultaneous loss alongside the loss of the penultimate segment in the male A1 in all nine species strongly supports its synapomorphic status.

Table 3. List of 39 morphological characters used for the here presented phylogenetic analysis. In the second column, plesiomorphic states are set in square brackets. Columns 3–14: 1 = apomorphies; 0 = plesiomorphies; ? = no information available; **1** = supposed convergences. 0* = also apomorphic state present, due to intraspecific variability; explanation in the text.

No.	Character [plesiomorphies in square brackets]/species	*L. sarsi*	*L. scottorum*	*L. volkerlehmanskii* sp. nov.	*L. georgei*	*L. gertraudae*	*L. spongiosus*	*L. mourois*	*L. typicus*	*L. monsmaris*	*L. macclintocki*	*L. sabinegeorgeae*	*L. whitsoni*
1	A2 exopod represented by tiny seta [with 1 small, knob-like segment bearing 1 small seta]	1	1	1	1	1	1	1	1	1	1	1	1
2	A1 male 6-segmented, chirocer [7-segmented, subcirocer]	?	1	1	1	1	1?	1	1	1	0?	0	0
3	P4 female enp-2 lacking outer seta [seta present]	1	1	1	1	1	1	1	1	1	0	0	0*
4	P3 exp-3 with at most 1 inner seta [with 2 setae]	1	1	1	1	1	1	1	1	0	**1**	0	0
5	P4 exp-3 with at most 1 inner seta [with 2 setae]	1	1	1	1	1	1	1	1	0	**1**	0	0
6	P2 exp-2 lacking inner seta [seta present]	1	1	1	1	1	1	1	1	0	0	0	0
7	P3 exp-2 lacking inner seta [seta present]	1	1	1	1	1	1	1	1	0	0	0	0
8	P2 exp-3 lacking inner seta [seta present]	1	1	1	1	1	1	0	0	0	0	0	0
9	P4 exp-3 inner apical seta trimmed down, flexible [seta almost identical with outer apical element]	1	1	1	1	0	0	0	0	0	0	0	0
10	P2 exp-3 inner apical seta trimmed down, flexible [seta almost identical with outer apical element]	1	1	1	0	0	0	0	0	0	0	0	0
11	P3 exp-3 inner apical seta trimmed down, flexible [seta almost identical with outer apical element]	1	1	1	0	0	0	0	0	0	0	0	0
12	P2 exp-3 down-trimmed inner apical seta bare [seta biplumose]	1	1	0	0	0	0	0	0	0	0	0	0
13	P3 exp-3 down-trimmed inner apical seta bare [seta biplumose]	1	1	0	0	0	0	0	0	0	0	0	0
14	P4 exp-3 down-trimmed inner apical seta bare [seta biplumose]	1	1	0	0	0	0	0	0	0	0	0	0
15	P5 female inner baseoendopodal seta of fish-bone aspect [seta bipinnate]	1	0	0	0	0	0	0	0	0	0	0	0
16	Body flattened [cylindrical]	0	1	0	0	0	0	0	0	0	0	0	0
17	Body somites laterally extended [not extended]	0	1	0	0	0	**1**	0	0	0	0	0	0
18	Anal operculum: posterior margin strongly serrated [with spinules]	0	0	1	0	0	0	0	0	0	0	0	0
19	Furcal tube pore long, displaced subapically [tube pore small, near furcal base]	0	0	1	0	0	0	0	0	0	0	0	0
20	P1 inner basal seta strongly diminished in size [of moderate length]	0	0	1	0	0	0	0	0	0	0	0	0
21	P1 outer basal seta with STE [lacking STE]	0	0	1	0	0	0	0	0	0	0	0	0
22	P1 exp-1 outer seta with STE [lacking STE]	0	0	1	0	0	0	0	0	0	0	0	0
23	P5 male exopod: subapical outer seta with STE [lacking STE]	0	0	1	0	0	0	0	0	0	0	0	0
24	P1 enp-2 apical long seta lost geniculation [seta geniculated]	0	0	0	1	0	0	0	0	0	0	0	0
25	P4 endopod 1 segmented [2-segmented]	0	0	0	1	0	0	0	0	0	0	0	0
26	P4 enp-2 lacking inner seta [seta present]	0	0	0	0	1	0	0	0	0	0	0	0*
27	Abdominal somites except telson dorsally with H-like cuticular structures [such structures absent]	0	0	0	0	0	1	0	0	0	0	0	0
28	P2 exp-3 innerapicalseta bare [seta biplumose]	0	0	0	0	0	0	1	1	0	0	0	0
29	P3 male enp-3 bulged out on its inner margin [margin straight]	0	0	0	0	0	0	1	0	0	0	0	0
30	Mxp lacking tiny seta accompanying claw [tiny seta present]	0	0	0	**?**	0	0	0	1	0	1	0	0
31	Mxp extremely strengthened [mxp of moderate size]	0	0	0	0	0	0	0	0	1	0	0	0
32	Telson overlapped by previous somite [not overlapped]	0	0	0	0	0	0	0	0	1	0	0	0
33	Rostrum frontally with tuft of long setules [no setular tuft]	0	0	0	0	0	0	0	0	0	0	1	0
34	Abdominal somites except telson dorsally with paired cuticular longitudinal ridges [such ridges absent]	0	0	0	0	0	0	0	0	0	0	1	0
35	Abdominal somites except telson dorsally with pairs of long tube pores [paired tube pores, if present, small]	0	0	0	0	0	0	0	0	0	0	1	0
36	FR mid-laterally with accessory long tube pore [lacking accessory tube pore]	0	0	0	0	0	0	0	0	0	0	1	0
37	FR setae I and II displaced subapically [arising mid-laterally]	0	0	0	0	0	0	0	0	0	0	1	0
38	P5 male exopod: proximal lateral seta with STE [lacking STE]	0	0	0	0	0	0	0	0	0	0	0	1
39	P5 male exopod: subapical inner seta with STE [lacking STE]	0	0	0	0	0	0	0	0	0	0	0	1

Remarks on character 2: Recent detailed descriptions of the male A1 revealed the existence of a very small fourth antennular segment between the third and the swollen fifth segment in *Laophontodes* (e.g., George and Gheerardyn 2015; George 2018). This tiny segment – already known for other Ancorabolidae – was first documented by Conroy-Dalton (2004) in males of other Laophontodinae (*Lobopleura ambiducta* Conroy-Dalton, 2004 and *Probosciphontodes* Fiers, 1988). The detection of a fourth antennular segment lead subsequent authors to confirm the presence of a small/tiny fourth segment in *Ancorabolina* George, 2006 (George 2006; George and Tiltack 2009; Gheerardyn and George 2010), *Bicorniphontodes* (George and Gheerardyn 2015; George et al. 2019), and *Calypsophontodes* Gheerardyn & Lee, 2012 (Gheerardyn and Lee 2012). This fourth segment has been also overlooked in *Laophontodes* (e.g., Schizas and Shirley 1994; Arroyo et al. 2003). However, the redescription of several laophontodin species (e.g., *Bicorniphontodes bicornis*, *Laophontodes typicus*, *L. whitsoni*), the description of new species of *Laophontodes* (George and Gheerardyn 2015; George 2018), and re-examination of available material of *Laophontodes mourois* (George pers. obs.) proved both the existence of this segment and that it had previously gone unnoticed. Therefore, it can be assumed with some certainty that this reduced fourth segment is also present in the male A1 of *L. macclintocki* and *L. spongiosus*.

Remarks on character 3: As documented by George and Gheerardyn (2015), the female P4 enp-2 in *L. whitsoni* apparently presents an intraspecific variability; three examined females had four setae – two apical, one outer and one inner (formula I:2:1) – , while other females lacked the outer seta (0:2:1) or even both lateral elements (0:2:0). Considering that the secondary development of a formerly deleted element is possible (cf. George 2020 and references therein) but rather improbable, we conclude that *L. whitsoni* originally bears all four elements in the P4 enp-2 (I:2:1). The reduction of the outer or both lateral setae is seen here as a deviation that has occurred within the species. Therefore, *L. whitsoni* is not grouped with those nine taxa that share the synapomorphic loss of the outer seta (character 3). Similarly, although *L. gertraudae* also lacks the inner seta of P4 enp-2 (character 26), a closer relationship with *L. whitsoni* cannot be presumed due to the rarity of this character in the latter species. Nonetheless, this intraspecific variation and potential relationships are indicated by an asterisk * in the respective fields in Table 3.

Further relationships between *Laophontodes macclintocki*, *L. sabinegeorgeae*, and *L. whitsoni* remain unsolved (Fig. 11). Whilst each of these species can be characterised by at least two autapomorphies (Table 3, Fig. 11), no derived characters have been found that might support any sister-group relationship.

Another four derived characters are shared by eight species (*L. sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., *L. georgei*, *L. gertraudae*, *L. spongiosus*, *L. mourois*, *L. typicus*; Table 3, Fig. 11):

4. P3 exp-3 with at most 1 inner seta [with 2 inner setae];
5. P4 exp-3 with at most 1 inner seta [with 2 inner setae];
6. P2 exp-2 lacking inner seta [seta present];
7. P3 exp-2 lacking inner seta [seta present].

Two inner setae on P3 exp-3 (character 4) and P4 exp-3 (character 5) are present in *L. monsmaris*, *L. sabinegeorgeae*, and *L. whitsoni*, while one inner seta was lost in the P3 and P4 exp-3 of the remaining *Laophontodes* species. This is seen as the derived state and thus as synapomorphic for the respective species. An exception is *L. macclintocki*, in which an inner seta is lost in the P3 and P4 exp-3. Unlike the other eight species in this group, *L. macclintocki* does not exhibit the synapomorphic state for characters 2 and 3, and therefore, the loss of the inner setae on the P3 and P3 exp-3 in *L. macclintocki* can be assumed to be convergent. The alternative would be to assume that the apomorphic character of the chirocer A1 is the result of convergent development, which is far more implausible.

Furthermore, the eight species share derived characters 6 and 7, viz. the loss of the inner seta on P2 and P3 exp-2, respectively. Although we admit that characters 6 and 7 are rather weak because the reduction of elements may occur independently (see remarks on character 3), it is assumed that, together with characters 4 and 5, they constitute a set of deviations that were developed in a common ancestor of the eight species (Table 3, Fig. 11) and are thus interpreted as synapomorphies for them.

Six species share a single derived character (*L. sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., *L. georgei*, *L. gertraudae*, *L. spongiosus*; Table 3, Fig. 11):

8. P2 exp-3 lacking inner seta [with 1 inner seta].

Among the above group of eight species, *Laophontodes typicus* and *L. mourois* (as well as all more basal species) show the plesiomorphic retention of an inner seta on the third exopodal segment of P2, whereas the remaining six species share its derived loss. This is seen here as synapomorphic for *L. spongiosus*, *L. gertraudae*, *L. georgei*, *L. volkerlehmanskii* sp. nov., *L. scottorum*, and *L. sarsi*.

Four species are characterised by the following putative synapomorphy (*L. sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., *L. georgei*; Table 3, Fig. 11):

9. P4 exp-3 inner apical seta short, flexible [seta of normal length].

In the harpacticoid ground pattern, the two apical setae of P2–P4 exp-3 are longer and more flexible than the outer spines of those segments, being of almost the same size. This state is retained in most *Laophontodes* species except for *Laophontodes sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., and *L. georgei*. These species are characterised by a clearly diminished inner apical seta of the P2–P4 exp-3, being much slenderer than the outer apical seta. This is interpreted as synapomorphic for these four species.

As with the other subgroups of the genus, relationships with those species excluded from the subgroup require further phylogenetic resolution (cf. interrogation marks in Fig. 11). The relationships of *Laophontodes spongiosus* and *L. gertraudae* with this last subgroup of four species, *L. sarsi*, *L. scottorum*, *L. volkerlehmanskii* sp. nov., and *L. georgei*, remain unclear, as no further apomorphic characters have been identified.

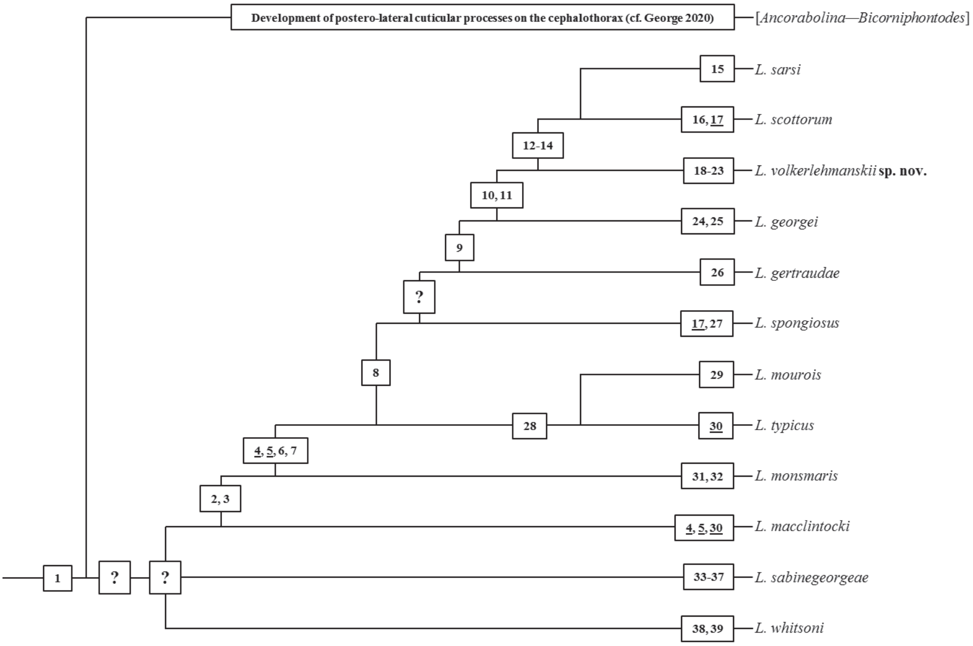

Figure 11. Cladogram summarizing the results of the phylogenetic analysis provided in the present contribution. Numbers in rectangles pointing to discussed characters listed in Table 3. Underlined numbers refer to convergent deviations. Detailed explanations are given in the text.

Three species – *L. sarsi*, *L. scottorum*, and *L. volkerlehmanskii* sp. nov. – share two further deviations (Table 3, Fig. 11):

10. P2 exp-3 inner apical seta short, flexible [seta of normal size];
11. P3 exp-3 inner apical seta short, flexible [seta of normal size].

In addition to the derived inner apical seta in the P4 exp-3, *Laophontodes sarsi*, *L. scottorum*, and *L. volkerlehmanskii* sp. nov. exhibit a short, flexible seta on the P2 and P3 exp-3, whilst *L. georgei* retains the normal-shaped inner apical setae. This is assumed as synapomorphic for the former species.

Finally, in *Laophontodes sarsi* and *L. scottorum* the inner apical seta of P2–P4 suffers a further deviation (Table 3, Fig. 11):

12. P2 exp-3 short inner apical seta bare [short inner apical seta biplumose];
13. P3 exp-3 short inner apical seta bare [short inner apical seta biplumose];
14. P4 exp-3 short inner apical seta bare [short inner apical seta biplumose].

In Harpacticoida, the inner and apical setae of P2–P4 exp-3 are usually biplumose, which must be regarded as the plesiomorphic condition. Thus, the development of unarmoured, bare setae constitutes a deviation. Accordingly, the presence of the bare, short seta in P2–P4 exp-3 is considered here as synapomorphic for *Laophontodes sarsi* and *L. scottorum*.

Remarks: The development of a bare inner apical seta in the P2 exp-3 is also present in *L. typicus* and *L. mourois* (Table 3, character 28). Nevertheless, we assume that the loss of the setal ornamentation occurred independently in these species. Shortening of the inner apical setae presumably took place before the loss of their armour in *Laophontodes sarsi* and *L. scottorum*. This assumption is further supported by the fact that these short setae remain biplumose in *L. volkerlehmanskii* sp. nov.. In contrast, the length of the inner apical seta in the P2 exp-3 of *L. typicus* and *L. mourois* is normal as in P3 and P4. A phylogenetic discussion of character 28 is given below.

The following characters, 15–39, listed in Table 3, characterise the different *Laophontodes* species. Direct comparison of characters was impossible for most species because of the lack of suitable type material. Consequently, the characterization of species by apomorphic characters is far from complete, with several species being characterised by just one potential apomorphy. Until further data are available, the current analysis provides sufficient information for an initial phylogenetic characterization of each species.

Characterization of *Laophontodes* species

Laophontodes sarsi, character 15 (Table 3, Fig. 11): The common shape of the harpacticoid P5 baseoendopodal setae (including *Laophontodes*) is a bipinnate one, with the pinnae being distinct. This is considered the plesiomorphic state. In contrast, the pinnae are strengthened and fused to the seta in *L. sarsi*, giving a "fish-bone" appearance (George 2018). This shape is rarely seen in Harpacticoida, and it is considered to be derived, i.e., an apomorphic state.

Laophontodes scottorum, characters 16 and 17 (Table 3, Fig. 11): George (2020) considered a cylindrical, slender body – listed here as character 1 in Table 3 – as synapomorphic for *Ancorabolina*, *Bicorniphontodes*, and *Laophontodes*. According to George (2020), the plesiomorphic state consists of a fusiform body that tapers posteriorly. *Laophontodes scottorum* deviates from character 1 in presenting a robust, rather compact body somewhat dorsoventrally compressed (character 16) (T. Scott 1907; George 2018). Moreover, the body somites are laterally extended (character 17); these lateral extensions are reminiscent of epimeres in other harpacticoid taxa (T. Scott 1907; George 2018). However, *L. scottorum* exhibits the synapomorphic state for characters 2–14, clearly justifying its assignment to *Laophontodes*. Moreover, although its body shape does not fit the synapomorphic state for *Laophontodes*, it does not match the plesiomorphic condition either. Instead, it can be postulated that the body shape of *L. scottorum* represents a secondary deviation, in addition to Character 17. Both character states are considered autapomorphic for this species.

Character 17 is also present in *L. spongiosus* (cf. Schizas and Shirley 1994); however, it only shares this character and apomorphies 2–8 with *L. scottorum*, suggesting it branched off much earlier, not sharing apomorphies 9–14. Therefore, we assume that the lateral extension of the body somites occurred convergently in these two species.

Laophontodes volkerlehmanskii sp. nov., characters 18–23 (Table 3, Fig. 11): This newly described species presents a series of morphological differences compared to the remaining species of *Laophontodes*. To characterise it unambiguously, the following six autapomorphies were selected: Strong serration of the posterior margin of the anal operculum (character 18), which is unique within *Laophontodes* – with the anal operculum of almost all other species exhibiting a row of fine spinules; subapical displacement and elongation of the furcal tube pore (character 19), compared to the usually small furcal tube pore located on the outer anterior lateral margin of the ramus in most *Laophontodes* species; a strongly diminished inner seta on the P1 basis (character 20), which does not reach the endopod in *L. volkerlehmanskii* sp. nov., similarly contrasts to the synapomorphic condition for *Laophontodes* in which the inner basal seta of the P1 is of moderate length, usually reaching the endopod; development of STE on the P1 outer basal seta (character 21), the P1 exp-1 outer spine (character 22), and the outer subapical seta of the male P5 exopod (character 23) are likewise exclusive derived features of *L. volkerlehmanskii* sp. nov., STE being rarely documented in Harpacticoida. In *Laophontodes*, only two species have been described possessing STE, namely *L. whitsoni* (characters 38, 39) (George and Gheerardyn 2015) and *L. volkerlehmanskii* sp. nov.. This has been confirmed by examination of various *Laophontodes* material (George pers. obs.). Thus, characters 18–23 are seen here as unambiguous autapomorphies of *Laophontodes volkerlehmanskii* sp. nov.

Laophontodes georgei, characters 24 and 25 (Table 3, Fig. 11): *L. georgei* was described as *L. norvegicus* George, 2018 by George (2018) and subsequently renamed by Lee and Huys (2019), with the illustrations provided by Sars (1908) as the holotype. It may be characterised by two deviations: Firstly, *L. georgei* has a long, non-geniculated apical seta on the P1 enp-2 (character 24) (Sars 1908) compared to a long geniculated seta in all other *Laophontodes* species, as well as in *Ancorabolina* and *Bicorniphontodes*, which are considered closely related. Consequently, the geniculated seta is considered to be the plesiomorphic state. As an early harpacticoid description, being more than 100 years old (Sars 1908; as *L. typicus*), one might suspect this geniculation was overlooked. However, G.O. Sars was a keen observer, and in fact noted geniculated setae on the P1 exp-3 of *L. georgei*. Thus, there is no reason to assume that he had overlooked the geniculation in the apical seta of P1 enp-2. It is therefore concluded that in *L. georgei* the P1 enp-2 apical seta lost the geniculation, resulting in an autapomorphic character for that species.

Moreover, *L. georgei* is the only *Laophontodes* species that exhibits a 1-segmented P4 endopod (Sars 1908; George 2018) (character 25). This reduction of the enp-1 is interpreted as autapomorphic of the species.

Laophontodes gertraudae, character 26 (Table 3, Fig. 11): All species of *Laophontodes*, except for *L. gertraudae*, bear a P4 enp-2 with 1 inner seta; only *L. gertraudae* lacks it (George 2018; but see discussion on character 3). This is regarded as an autapomorphy for the species.

Laophontodes spongiosus, characters 17 and 27 (Table 3, Fig. 11): *L. spongiosus* has three derived characters that are pooled as one autapomorphy (character

27): the abdominal somites except the telson are characterised by H-like cuticular dorsal structures (Schizas and Shirley 1994). Such structures are unique within *Laophontodes*. They are reminiscent of similar structures found in *Paralaophontodes* (Lang 1965; Fiers 1986; George 2017), but as shown by George (2020), no closer relationship between *Paralaophontodes* and *L. spongiosus* exists. So, although these derived features appear to be convergent for the two taxa, they are considered autapomorphic for the latter.

In addition to character 27, another deviation discussed here is interpreted as convergent (character 17, cf. *L. scottorum*).

Laophontodes mourois–L. typicus-group, character 28 (Table 3, Fig. 11): The transformation of pinnate/plumose setae into bare elements has been discussed above (character 12). The rather ancestral inner apical seta in the P2–P4 exp-3 of *Laophontodes* is biplumose, as observed in, for example, *L. gertraudae*, *L. monsmaris*, *L. spongiosus*, and *L. whitsoni*. Within the genus, however, two developmental directions were detected. The first is the reduction in length of the inner apical setae, followed by a subsequent loss of ornamentation (characters 9–11; 12–14); this is seen in *L. georgei* (on the P2), and in *L. volkerlehmanskii* sp. nov., *L. scottorum*, and *L. sarsi* (on P2–P4), and has been discussed above. A second developmental direction is seen in *Laophontodes mourois* and *L. typicus*, in which the length of the inner apical seta of the P2 exp-3 is normal, but has lost it ornamentation. This derived state is considered as synapomorphic for *Laophontodes mourois* and *L. typicus*.

Laophontodes mourois, character 29 (Table 3, Fig. 11): Based on the description of Arroyo et al. (2003), *L. mourois* has one autapomorphy: The male P3 enp-3 shows a rounded inner margin (character 29) compared to other *Laophontodes* males with a straight inner margin.

Laophontodes typicus, character 30 (Table 3, Fig. 11): *L. typicus* does not present any exclusive morphological deviations. Compared with other *Laophontodes* species, *L. typicus* seems to retain most plesiomorphic character states. Only two deviations have been observed in the species, characters 28 and 30, and these are shared with other congeners. Of these, character 28 supports a sister-group-relationship with *L. mourois* (see above). In contrast, character 30 – the lack of the minute seta accompanying the maxillipedal claw – , whilst also found in *L. macclintocki* (see below), is thought to be the result of convergence: *L. macclintocki* lacks the apomorphic state of character 28, but exhibits apomorphies 4 and 5 (see below), which are not seen in *L. typicus*. Therefore character 30 is regarded as autapomorphic for *L. typicus*.

According to the description of Sars (1908), no such minute seta is present in the maxillipedal claw of *L. georgei*. Future examination may reveal if this is true or if the seta was overlooked by Sars (1908).

Laophontodes monsmaris, characters 31, 32 (Table 3, Fig. 11): This species exhibits two autapomorphic characters, which are unique not only within *Laophontodes* but also in the Laophontodinae: the maxilliped is extremely elongated and strengthened (character 31), and the penultimate abdominal somite overlaps the telson (character 32) (George 2018). Because of these autapomorphies a phylogenetic characterization of *L. monsmaris* is unambiguous.

Laophontodes macclintocki, characters 4, 5, 30 (Table 3, Fig. 11): The convergent loss of 1 inner seta in P3 and P4 exp-3 (characters 4 and 5) has been discussed above. In addition, *L. macclintocki* shares one further (convergent) deviation with *L. typicus*, viz. the loss of the tiny seta accompanying the maxillipedal claw (character 30). As stated by George (2018), the loss of this seta must be considered with care, since it has been overlooked in species descriptions. However, with respect to *L. macclintocki* we trust in the description of Schizas and Shirley (1994), who noted this tiny seta in *L. spongiosus* in the same publication and are therefore unlike to have missed it in *L. macclintocki*. As discussed above, we hypothesise that the loss of this seta is autapomorphic for *L. macclintocki* and that its absence in *L. typicus* is the result of convergence.

Laophontodes sabinegeorgeae, characters 33–37 (Table 3, Fig. 11): *L. sabinegeorgeae* may be unambiguously characterised by several derived characters (cf. George 2018). The species exclusively presents a tuft of long setules on the front of the rostrum (character 33); the presence of paired longitudinal cuticular ridges on the abdominal somites except the telson (character 34); the development of paired, remarkably long tube pores on the abdominal somites except the telson (character 35); a long tube pore arising mid-laterally on the FR (character 36), in addition to a small anterior tube pore found on the FR in other *Laophontodes* species, and, finally, the subapical displacement of furcal setae I and II (character 37).

Remarks on character 37: According to George et al. (2019) and Lee and Huys (2019), furcal setae I and II in species of *Laophontodes* are positioned in the distal half of the furcal rami, close to the centre line. This is the case in nine species, *L. georgei*, *L. gertraudae*, *L. monsmaris*, *L. mourois*, *L. sarsi*, *L. scottorum*, *L. typicus*, *L. volkerlehmanskii* sp. nov., and *L. whitsoni* (cf. Sars 1908; Arroyo et al. 2003; George and Gheerardyn 2015; George 2018; present contribution, Fig. 5A). However, a trend towards the apical displacement of setae I and II can be noted: in *L. macclintocki* and *L. spongiosus* they are displaced distally but still positioned on the outer lateral margin of the FR; in *L. sabinegeorgeae* they are almost in the subapical margin of the FR (George and Gheerardyn 2015). This latter position also resembles the derived condition as found in *Bicorniphontodes* (cf. George et al. 2019). The subapical position of furcal setae I and II in *L. sabinegeorgeae* is unique in *Laophontodes*.

Laophontodes whitsoni, characters 38 and 39 (Table 3, Fig. 11): *L. whitsoni* is the first species that branches off in the cladogram presented in Fig. 11. It presents two deviations regarding the male P5: both the proximal outer seta (character 38) and the subapical inner seta (character 39) present STE (George and Gheerardyn 2015) which are absent in all remaining *Laophontodes* species.

Remarks: Five species (*L. whitsoni*, *L. sabinegeorgeae*, *L. typicus*, *L. mourois*, and *L. scottorum*) present a further character that must be regarded as deviation, that is the development of fine longitudinal ripples dorsally on the pedigerous somites bearing the P2–P4 (Arroyo et al. 2003; George and Gheerardyn 2015; George 2018). Such ripples may even be seen on the remaining pedigerous somites as well as on the abdominal somites (except telson). However, if this character state is to be considered as synapomorphic for the above species, this would demote characters 2–14 to convergences (Table 3), which would be less parsimonious. Furthermore, while the absence of such ripples is confirmed for *L. gertraudae*, *L. monsmaris*, and *L. volkerlehmanskii* sp. nov. (George 2018; present contribution), it is still not known if they occur in *L. antarcticus*, *L. georgei*, *L. macclintocki*, *L. propinquus sp. inquirenda*, and *L. spongiosus*. Consequently, it was not possible to include this character in the here presented study.

Summary and conclusion

The description of *Laophontodes volkerlehmanskii* sp. nov. facilitated an attempt to characterise the genus *Laophontodes* and to elucidate the phylogenetic relationships within the taxon. Careful comparison of 39 morphological characters led to the conclusion that *Laophontodes* cannot be characterised by any autapomorphies. Instead, it seems to reflect the stem-lineage of a monophylum comprised of *Ancorabolina*, *Bicorniphontodes*, and *Laophontodes*. While *Ancorabolina* and *Bicorniphontodes* can be characterised as monophyla and furthermore present a sister-group relationship (George 2020), *Laophontodes* retains the characters of the common ancestor, without having developed unique deviations that might be considered as synapomorphies of species assigned to the genus.

Similarly, discrimination of the 12 *Laophontodes* species examined here (*L. antarcticus* and *L. propinquus* excluded) is ambiguous. Most characters refer to the reduction of single setae or spines, which happens often and independently in harpacticoid species. Moreover, several features presumed to be derived, such as the development of fine dorsal cuticular ripples on the pedigerous somites P2–P4, or the lateral extension of the body somites, seem to be distributed quite heterogeneously amongst the species. Finally, many descriptions of *Laophontodes* species are incomplete or of poor quality, and the respective type material is no longer available. Those conditions have inhibited the comparison of all the morphological characters that may be otherwise of phylogenetic relevance.

Nonetheless, each of the *Laophontodes* species can be characterised by certain derived characters, even if convergence has to be assumed for some of them. Thus, the phylogenetic analysis undertaken provides insights into the phylogenetic relationships of and within *Laophontodes* and serves as the base for ongoing research.

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Review of the mite genus *Krantzolaspina* Datta & Bhattacharjee (Mesostigmata, Parholaspididae) with re-description of *K. angustatus* comb. nov. (Ishikawa) from Indonesia

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Abstract

Herein, we update the diagnosis and description of the genus *Krantzolaspina* Datta & Bhattacharjee and provide a list of the three valid species including new combinations and synonyms, as follows: 1) *Krantzolaspina angustatus* (Ishikawa, 1987) **comb. nov.** (= *Indutolaelaps jiroftensis* Hajizadeh et al., 2017 **syn. nov**.), 2) *K. rebatii* Datta & Bhattacharjee, 1989 and 3) *K. solimani* (Metwali, 1983) **comb. nov.** Finally, we re-describe *K. angustatus* (Ishikawa, 1987) **comb. nov.** based on the holotype from Japan, voucher specimens from Iran and additional females that we found in soil samples from oil palm plantations in Sumatra, Indonesia.

Keywords

Gamasina, monotype, morphology, oriental region, rainforest, tropical lowland

Introduction

The mesostigmatic mite family Parholaspididae has a total of 14 known genera distributed worldwide (Marchenko 2016). Species of Parholaspididae are found in a variety of habitats, such as soil-litter of forests and disturbed habitats (Jung et al. 2010), dead wood, moss, tree holes and some have been found associated with mammal nests, such as Cricetidae and Muridae (Yin et al. 1964, Petrova 1967a, 1967b, Gu 1984, Chen et al. 1994, Gu and Guo 1996). Other species have been reported from caves (Willmann 1940, Ishikawa 1995a, 2002) and also as early colonisers after habitat disturbance.

Indonesia is a biodiversity hotspot (Rintelen et al. 2017). However, knowledge on most groups of mites is still very limited. For instance, the family Parholaspididae is represented by only two described species (*Holaspulus viduus* Berlese, 1905 and *Parholaspis desertus* Berlese, 1918; both from Java). This is comparatively low with some countries in the Oriental region that have many more species records of Parholaspididae, such as China (56 species), Japan (28), Philippines (9) and Malaysia (7) (Berlese 1905, 1918, Vitzthum 1926, Evans 1956, Krantz 1960, Yin et al. 1964, 1999, Ishikawa 1966, 1969, 1976, 1979, 1980a, 1980b, 1987a, 1987b, 1993, 1994, 1995a, 1995b, 2002, Petrova and Tascaeva 1968, Bhattacharyya 1977, Gu 1984, Datta and Bhattacharjee 1989, Liang and Hu 1993, Tseng 1993, Yin and Bei 1993, Chen et al. 1994, Lee and Cho 1995, Gu and Guo 1996, Lee and Lee 1996a, 1996b, 2000, Ishikawa and Saichuae 1997, Ma 1998, 2004, 2010, 2012, Ma and Yin 1999, Ma and Yan 2001, Bei et al. 2004, 2009, Ma and Lin 2006, 2011, 2013, 2014a, 2014b, 2015, Bai and Ma 2014, Bai et al. 2014, Lee 2014, Kontschaán et al. 2015, Bhattacharyya and Kheto 2016). Other countries in the region have few species recorded but that may merely indicate a lack of knowledge: India (3 species), the Korean peninsula (1) and Thailand (1).

Despite the remarkable faunal diversity of the oriental region and numerous reported species of parholaspidid mites, there are shortcomings in the old species descriptions (lack of leg chaetotaxy, information on external poroidotaxy and adenotaxy of the idiosoma). For the previous, we decided to add information and organised the monotypic genus *Krantzolaspina,* based on available specimens, photographs from paratypes, original description and illustrations. For this reason, in the present work, a re-description of *Krantzolaspina angustatus* comb. nov. based on holotype, review of the generic concept based on type material and literature, a new synonymy and a list of valid species with some comments is presented.

Material and methods

This study forms part of an investigation on soil and canopy arthropods of rainforests and agricultural systems in Jambi Province, Sumatra, Indonesia and was conducted within the framework of the interdisciplinary project "Ecological and socioeconomic functions of tropical lowland rainforest transformation systems (Sumatra, Indonesia)"

- EFForTS. For more details on the study region and the experimental design of the project see Drescher et al. (2016).

Mites were extracted from soil of oil palm plantations in the vicinity of Bukit Duabelas National Park, Jambi Province, Sumatra, Indonesia. Soil samples (1–3 cm depth) were taken using a spade and each consisted of a core of 16 × 16 cm area. Mites were extracted from samples using the high gradient canister method described in Kempson et al. (1963). All specimens were collected in November 2013 by B. Klarner. Mites were stored in 70% ethanol until slide-mounting in Hoyer's medium. For each mite, the gnathosoma was separated from the idiosoma and mounted next to it on the same slide.

Photographs and measurements were made using an Axiolab 5 phase contrast Zeiss microscope with an Axiocam 105 HD digital camera and Nikon Eclipse Ci connected to a computer-controlled digital camera Sight Ds-L3. Stacks of images were taken for each mite, using manual control of the focal plane. Selected images were combined using Zerene Stacker, version 1.04 (Zerene Systems, LLC 2009-2014). In some cases, images captured from different regions of the body were combined using the 'photomerge' function in Adobe Photoshop, version 2015 (16.0 or 20150529.r.88; Adobe Systems Inc., San Jose, USA). Digital drawings were prepared with Adobe Illustrator, version CC 2015 (19.0.0), based on (combined) photographs.

All measurements are given in micrometres (µm) and include the range (minimum–maximum). Lengths of shields were measured along their midlines and widths at their widest point, except for the sternometasternal shield which was measured at the level of insertion of setae *st2* and genitiventrianal shield between bases of *JV1–2*. Leg measurements were taken from the proximal margin of the coxa, along the midline of each segment, to the tip of the claw. Notations of body structures and idiosomal chaetotaxy follow Lindquist and Evans (1965) as adapted by Moraza and Peña (2006) and Marchenko (2016). Leg chaetotaxy follows Evans (1963) and Evans and Till (1965) and palps Evans and Till (1963). Idiosomal and peritrematal shield notations for pore-like structures (gland pores and poroids/lyrifissures) follow the system of Athias-Henriot (1971) for the ventral idiosoma and Athias-Henriot (1975) for the dorsal idiosoma.

Specimens of examined *K. angustatus* comb. nov. are deposited at NSMT (National Science Museum Natural History), Tokyo, Japan, the Holotype and voucher specimens at ESALQ (Escola Superior de Agricultura Luiz de Queiroz - Universidade de São Paulo), São Paulo, Brazil. Other collected materials are deposited at LIPI (Indonesian Institute of Science), Cibinong, Indonesia; SMNG (Senckenberg Museum), Görlitz, Germany; OSAL (Ohio State Acarology Collection), Columbus, USA and in ANIC (Australian National Insect Collection) Canberra, Australia. Additional photos of the species are digitally deposited in the online database Ecotaxonomy, accessible at http://www.ecotaxonomy.org.

The updated diagnosis and description of the genus were prepared after consulting the original description of the genus *Krantzolaspina* (Datta and Bhattacharjee 1989), as well as species descriptions (Metwali 1983, Ishikawa 1987a, Datta and Bhattacharjee 1989, Hajizadeh et al. 2017).

Taxonomic accounts

Parholaspididae

Genus *Krantzolaspina* Datta & Bhattacharjee

Krantzolaspina Datta & Bhattacharjee, 1989: 411.

Type species. *Krantzolaspina rebatii* Datta & Bhattacharjee, 1989.

Diagnosis. ***Female.*** Dorsal shield entire, usually bearing 32–36 pairs of setae. Presternal area with two pairs of free presternal platelets. Sternometasternal shield well-defined, bearing four pairs of setae. Genitiventrianal shield, bearing four pairs of setae: one pair of genital setae *st5* and three pairs of preanal setae (*Zv1* and *Jv1–2*) in addition to circumanal setae. Podal-peritrematal shield free from genitiventrianal shield. Epistome with long median projection and with lateral margins serrate. Cheliceral movable digit with a pair of unequal plumose arthrodial brushes and no arthrodial corona or with a single plumose arthrodial brush and a fringed arthrodial corona. Palp trochanter with a small spur-like process near its ventral base (not palpfemur as in the original description); palptarsal claw three-tined. Pretarsus I reduced or absent, pretarsi II–IV well-developed. Chaetotaxy of femur-genu-tibia of legs I: 13-12-12; II: 11-11-10; III: 6-8-8; IV: 6-8-8.

Male. Unknown.

Description. ***Female.*** *Dorsal idiosoma.* Dorsal shield 340–561 µm long, broad anteriorly, gradually tapering posteriorly, anterior margin almost straight/truncate, partially covering idiosoma, leaving with the lateral and posterior margins of the soft integument broadly or narrowly exposed; shield mostly reticulate. Dorsal shield hypotrichous, bearing 32 (*J5* and *S5* slightly pilose in *K. angustatus* comb. nov.) or 36 pairs of smooth setae, most setae long and of similar length. Unsclerotised cuticle with 10–15 pairs of *r*, *R* and/or *UR* setae combined, smooth and moderately long (*UR* setae slightly pilose in *K. angustatus*).

Ventral idiosoma. All setae aciculate, smooth and relatively long (except *Jv4–5*, *Zv4–5* pilose in *K. angustatus*). Tritosternum with a pair of free pilose laciniae. Presternal area with two pairs of free, well-sclerotised presternal platelets. Sternometasternal shield well defined, longer than wide. Shield with posterior and anterior margin concave, partially smooth, reticulate or punctate (anterior and lateral margins ornamented in *K. angustatus*), bearing four pairs of setae *st1–4* and three pairs of poroids *iv1–3*. Peritrematal shield well-developed, broad, anteriorly free and posteriorly fused with developed podal shield; two pairs of poroids (*id7*, *ip*) and one pair of gland pores (*gdp*) posterior to stigma; Genitiventrianal shield longer than wide, reticulate, flask-shaped; cribrum developed. Soft opisthogastric cuticle with 6–7 pairs of setae; never hypertrichous. Metapodal platelets present or absent.

Gnathosoma. Subcapitulum with the corniculi well sclerotised, elongated and horn-like. Internal malae bifurcate, well separated from each other, densely fimbriated on outer margin and with apices slightly shorter than corniculi. Deutosternal groove with multidentate transverse rows, subcapitular setae smooth and aciculate, *h1–h3* often longer than *h2* and *pc*. Surface posterolaterad to seta *pc* with a pointed spine-like process or absent (simply flat). Epistome with a median projection and lateral margins irregularly serrate or may be smooth. Chelicera chelate-dentate; movable digit often bearing two teeth. A setiform pilus dentilus; smooth dorsal cheliceral seta, dorsal lyrifissure, a pair of unequal plumose arthrodial brushes or one plumose arthrodial brush and a narrow fringed arthrodial corona at base of the movable digit are present. Palp trochanter with a small pointed spine-like process in the ventral surface and with setae *v1* and *v2* slightly thickened and long, in contrast with setae of the other palp segments; palp tarsal claw three-tined.

Legs chaetotaxy as in diagnosis (see above).

Male. Unknown.

Re-description

Krantzolaspina angustatus (Ishikawa, 1987) comb. nov.

Proparholaspulus angustatus– Ishikawa, 1987a: 82; Kadkhodae et al., 2013: 131.
Indutolaelaps jiroftensis Hajizadeh et al., 2017 **syn. nov.**

Diagnosis. *Female.* Dorsal shield entire, mostly reticulate (except the anteromedial region which is smooth), partially covering the idiosoma, broad anteriorly and posteriorly narrowing; shield bearing 32 pairs of setae, most setae moderately long and smooth, except *j1–2* and *z1* which are slightly shorter and setae *S5* and *Z5* slightly pilose. Unsclerotised lateral cuticle of the idiosoma with a total of 15 pairs of smooth setae of similar length, except five pairs of *UR* setae slightly pilose, the pairs of setae *r6* and *R1* slightly shorter than *r5–7* plus seven pairs of *UR* setae. In the idiosoma ventre, all setae aciculate and smooth, except *JV4–5* and *ZV4–5* which are pilose. Presternal area with two pairs of free and presternal platelets. Sternometasternal shield mostly reticulate and covered by semi-rounded cells in the lateral margins, bearing four pairs of setae. Peritrematal shield anteriorly free, fused to the sternometasternal shield between coxae I–III, posteriorly fused with the parapodal shield and extended beyond posterior margin of the coxae IV, region of peritrematal + parapodal shield reticulate and covered by some semi-rounded cells; peritreme extending between coxae I–II at level of *st1*. Genitiventrianal shield longer than wide, reticulate, flask-shaped and bearing four pairs of setae *st5*, *JV1–3,* as well as three circumanal setae *pa* and *po*. Soft opisthogastric ventral cuticle with six pairs of setae *JV4–5*, *ZV2–5*. Metapodal platelets free, small and elliptical in shape. Deutosternal groove with six transverse rows, surface of the gnathosoma with pointed spine-like process similar to the ventral surface of the palp trochanter. Epistome with median projection bifurcate or trifurcate distally, lateral edges finely and irregularly serrate. Cheliceral digit movable and fixed with four and two teeth, respectively, base of movable digit with a plumose arthrodial brush and a narrow fringed arthrodial corona. Palp tarsal claw three-tined. Pretarsus I reduced or absent and pretarsi II–IV with pretarsi well-developed.

Male. Unknown.

Female (Figs 1–6) (*n* = *6*). *Idiosomal dorsum* (Fig. 1). 541–611 µm long, 287–372 µm wide. **Dorsal shield** 514–536 µm long, 219–239 µm wide (at level between *r2–3*), entire and broad anteriorly with the anterior margin slightly straight and posteriorly narrowing, covering partially the idiosoma with the lateral and posterior margins of soft integument broadly exposed, most surface of the shield reticulate, but smooth on the dorsocentral region between the bases of *z1* and *J2*. Dorsal shield bearing 32 pairs of setae of similar length and shape, most setae relatively long (36–46 µm) and smooth, except *j1–2* and *z1* slightly shorter (31–35 µm) and the setae *S5* and *Z5* slightly pilose (Fig. 1). **Podonotal region** with 17 pairs of setae *j1–6*, *z1–2*, *z4–6*, *s2*, *s6*, *r2–5* and 10 pairs of poroids, including three pairs of glands *gd1–2* and *gd4*. **Opisthonotal region** with 15 pairs of setae *J1–5*, *Z1–5*, *S1–5* and 10 pairs of poroids, including two glands *gd8* and *gd9*. **Unsclerotised lateral cuticle** bearing a total of 15 pairs of smooth setae of similar length (38–44 µm) except five pairs of *UR* setae slightly pilose (Fig. 1); the pairs of setae *r6* and *R1* slightly shorter than *r5–7* and the six pairs of *UR* setae (two of them on the ventral side); a pair of lyrifissures (*Rp*) present between *R3* and *R4*.

Idiosomal venter (Figs 2, 3A–C). All setae aciculate and smooth, except *Jv4–5*, *Zv4–5* which are pilose (Fig. 2). **Tritosternum** (Fig. 2) with columnar base and pair of free pilose laciniae. **Presternal area** bearing two pairs of free, well-sclerotised, transversely aligned and presternal platelets *ppl* (Figs 2, 3A–C), with transversal lineae over surface. **Sternometasternal shield** (Figs 2, 3A–C) 202–209 µm long, 104–110 µm wide (at level of *st2*) well-defined, reticulate and covered by semi-rounded cells in the lateral margins, posteriomedial area smooth beyond to the setae *st3*; anterior and posterior shield margin concave, bearing four pairs of setae *st1–st4* (36–41) and three pairs *iv1–3* of slit-like poroids; *iv1* larger than *iv2–3*, the latter rounded. **Peritrematal shield** (Fig. 2) broad, anteriorly free, fused to the exopodal shield (distinctly more sclerotised), to sternometasternal shield between the coxae I–III and posteriorly with the well-developed parapodal shield; shield extended beyond the posterior margin of the coxa IV, this area reticulate and covered by some semi-rounded cells; two pairs of poroids (*id7*, *ip*) and one pair of gland pores (*gdp*) discernible. Peritreme extending anteriorly between coxae I–II, at level of seta *st1*. **Genitiventrianal shield** (Figs 2, 3A–C) 246–264 µm long × 127–135 µm wide, reticulate and flask-shaped; shield with four pairs of setae *st5* 35–37 (slit-like poroids *iv5* on unsclerotised cuticle and posterolaterad of *st5*) *ZV1*, *JV1–2* 39–42 additionally to the circumanal setae *pa* and *po*, paranal setae *po* (27–29) aligned with anterior margin of anal opening, postanal seta shorter *po* (15–17); gland opening *gv3* on posterolaterad shield margins, at level slightly anterior to paranal setae, cribrum well-developed. Soft opisthogastric cuticle with seven pairs of setae *JV3–5*, *ZV2–5* (39–40) and three poroids, including one (*ivp*). **Metapodal** (Fig. 2; *met*) platelets free, small and suboval.

Gnathosoma (Fig. 4A–L). **Subcapitulum** (Fig. 4A–C): corniculi well sclerotised, elongated and horn-like slightly shorter than cheliceral movable digit. Internal malae bifurcate, totally separated from each other, densely fimbriated on outer margin and with apices slightly shorter than corniculi. Deutosternal groove with six transverse rows denticles, each bearing 8–21 denticles and distal row smooth, with lateral ridges each side

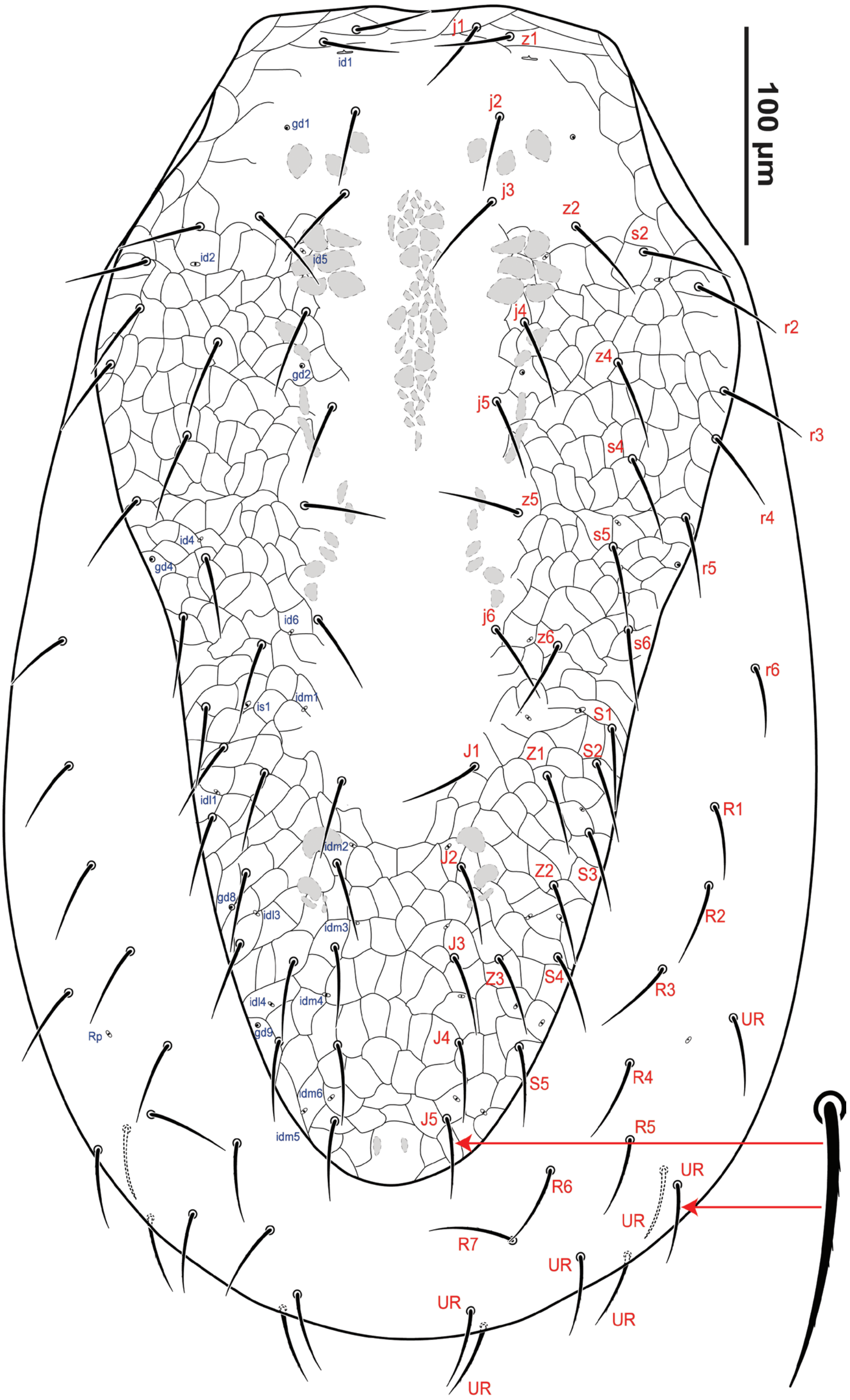

Figure 1. *Krantzolaspina angustatus* comb. nov., adult female. Dorsal idiosoma.

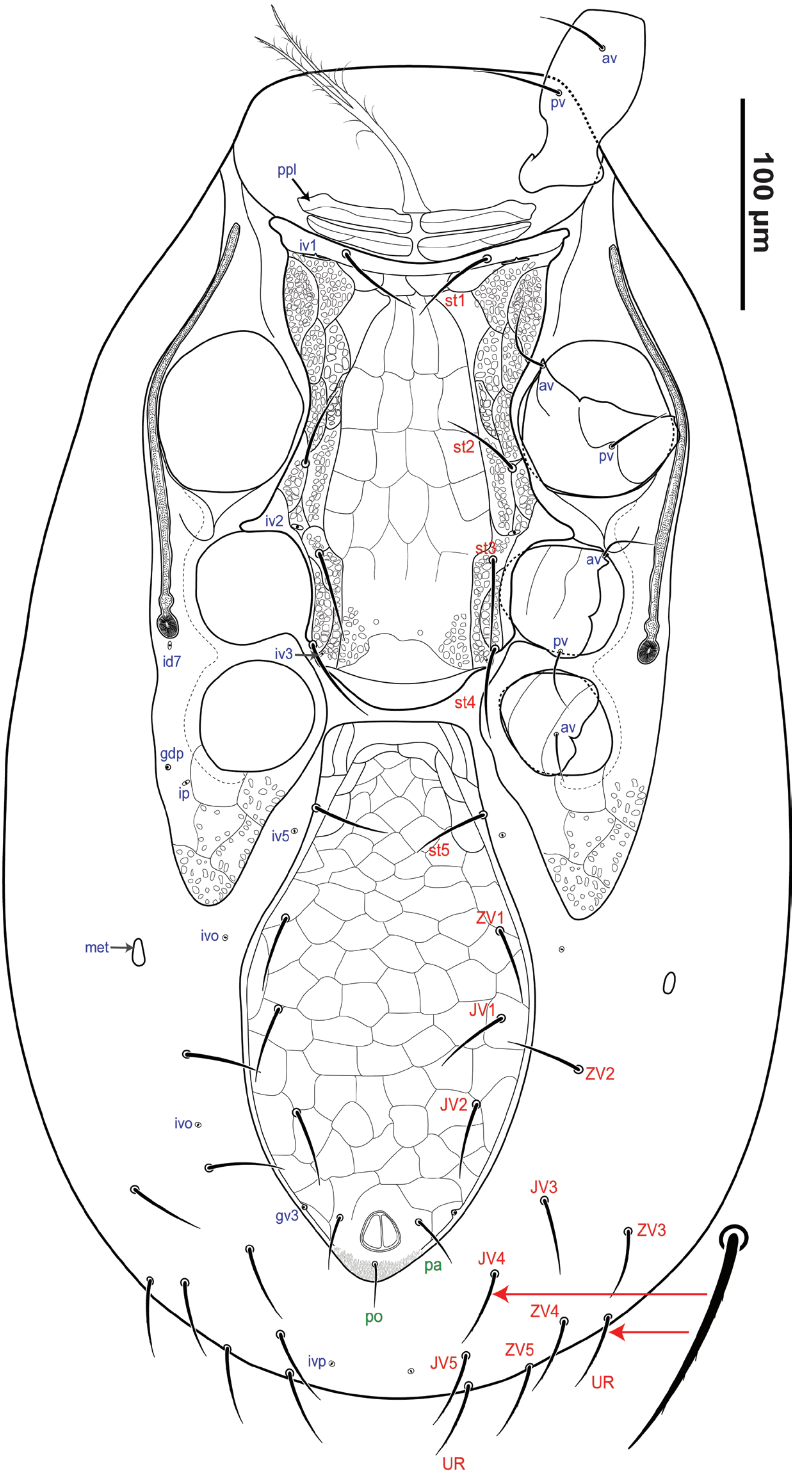

Figure 2. *Krantzolaspina angustatus* comb. nov., adult female. Ventral idiosoma.

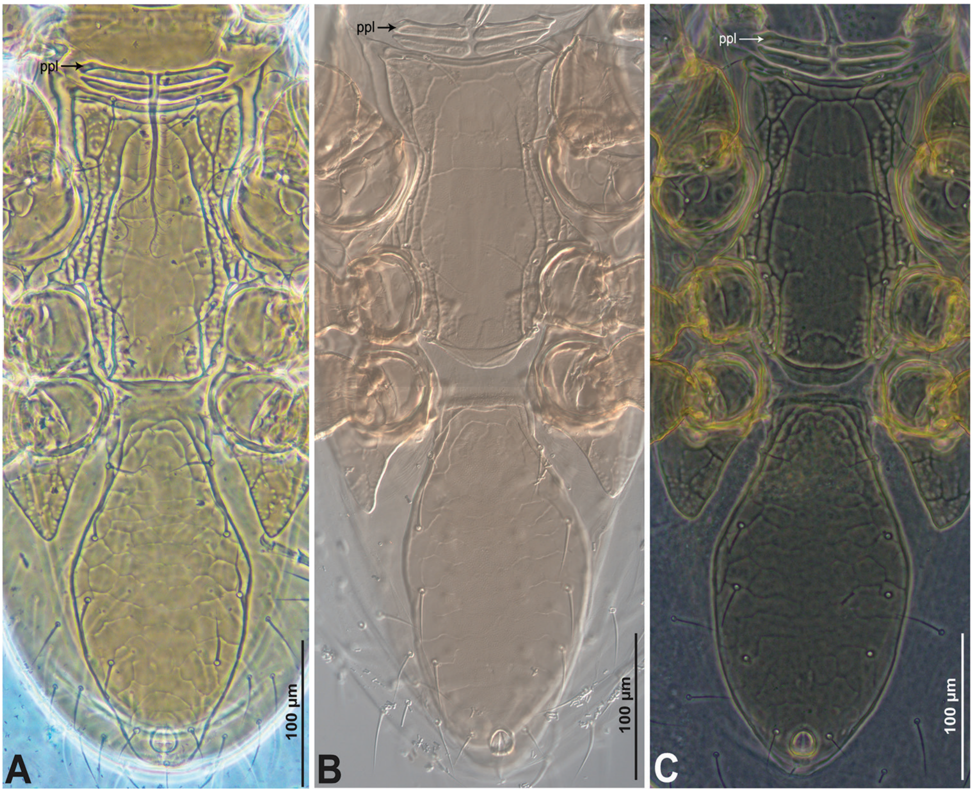

Figure 3. *Krantzolaspina angustatus* comb. nov., adult female. Ventral idiosoma showing the sternometasternal, genitiventrianal and parapodal shield. **A** Holotype of *Krantzolaspina angustatus* **B** Paratype of *Indutolaelaps jiroftensis* syn. nov., photos by Raphael Castilho **C** New material from Indonesia.

of the 2nd and 3rd row; subcapitular setae smooth and aciculate, *h1* 51–54 and *h3* 40–43 longer than *h2* 34–37 and *pc* 17–20. Surface posterolaterad to seta *pc* with a minute or a small spine-like process (Fig. 4A–C). Epistome (Fig. 4D–F) with a median projection bifurcate or trifurcate distally, lateral edges finely and irregularly serrate. **Cheliceral** (Fig. 4G–I) fixed digit (106–111) with four teeth (most proximal small and most distal inserted subapically) plus one distal hook-like tooth and a setiform *pilus dentilus*; dorsal and anti-axial lyrifissures present as well as dorsal setae smooth; movable digit (97–103) with two teeth directed backwards and one distal hook-like tooth, base of the movable digit with a plumose arthrodial brush (44–48) (Fig. 4I, see arrow) much shorter than movable digit and a narrow fringed arthrodial corona which is only discernible ventrally. **Palp** (Fig. 5) with normal chaetotaxy for Parholaspididae, with 2-5-6-14-15 setae on trochanter-femur-genu-tibia-tarsus; palptrochanter almost twice longer than palpfemur, with a small pointed spine-like process on the ventral surface (Figs 4J–L, 5; see arrow) and setae *v1*–2 thickened and long, in contrast to the setae of the other palp segments; palpfemur *al* thickened, palpgenu with setae *al1*–2 thickened and spatulate distally. Palp tarsal claw three-tined, medial tine conspicuous distally spatulate.

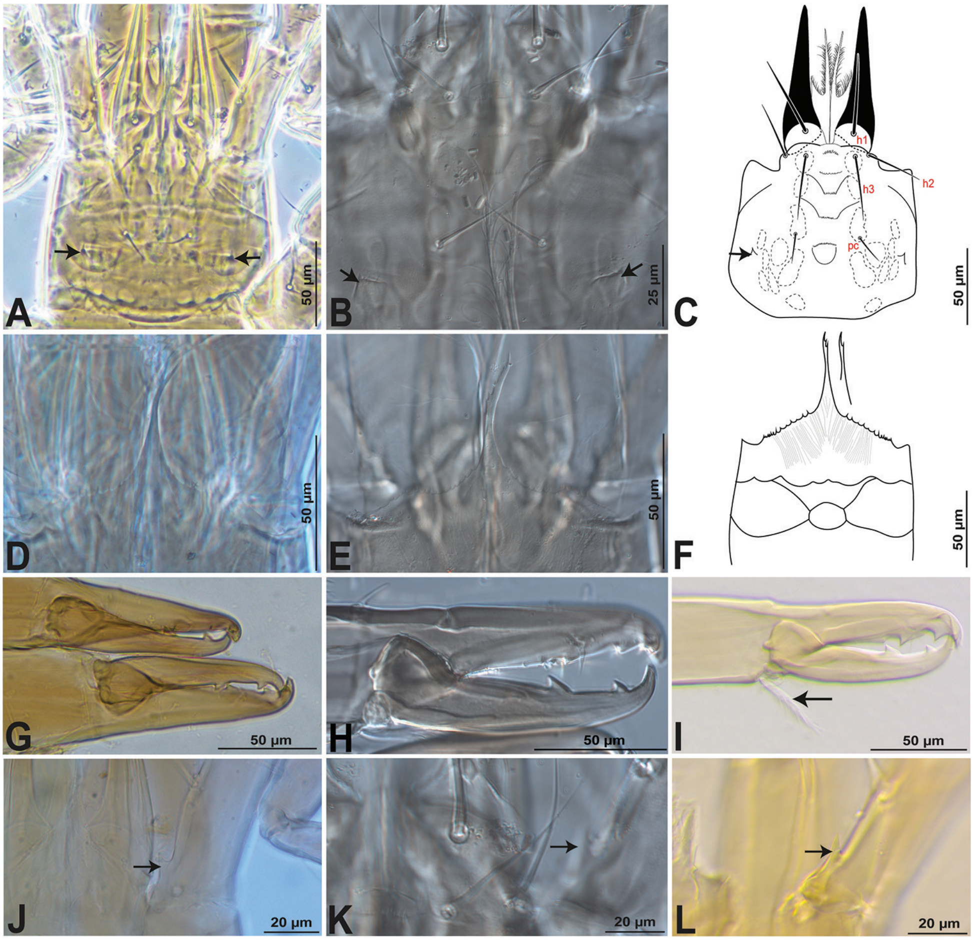

Figure 4. *Krantzolaspina angustatus* comb. nov., adult female, Gnathosomal structures. Holotype (Left); *Indutolaelaps jiroftensis* syn. nov. (Middle), photos by Raphael Castilho; New material from Indonesia (Right). **A–C** Subcapitulum **D–F** Epistome **G–I** Chelicera **J–L** Palp trochanter with a ventral spine-like process.

Legs (Fig. 6A–D) lengths (in µm): I (Fig. 6A): 573–625, II (Fig. 6B) 461–495, III (Fig. 6C) 396–421, IV (Fig. 6D) 550–575. The leg chaetotaxy/setation—Coxae: I 2 (0, 0/1, 0/1, 0); II: 2 (0, 0/1, 0/1, 0); III: 2 (0, 0/1, 0/1, 0); IV: 1 (0, 0/0, 0/1, 0); trochanters: I: 6 (1, 0/1,1/2, 1); II: 5 (1, 0/1, 0/2, 1); III: 5 (1, 1/1, 0/2, 0); IV: 5 (1, 1/1, 0/2, 0); femora: I: 13 (2, 3/1, 2/3, 2); II: 11 (2, 3/1, 2/2, 1); III: 6 (1, 2/1, 1/0, 1): IV: 6 (1, 2/1, 1/0, 1); genua: I: 12 (2, 3/2, 2/1, 2); II: 10(2, 2/1, 2/1, 2); III: 8 (2, 2/1, 2/1, 1); IV: 8 (2, 2/1, 2/0, 1); tibia: I: 12 (2, 3/2, 2/1, 2); II: 10 (2, 2/1, 2/1, 2); III: 8 (2, 1/1, 2/1, 1); IV: 8 (2, 1/1, 2/1, 1); tarsi I: not counted, II: 18; III: 18; IV: 18. Pretarsus I reduced or absent and pretarsi II–IV well-developed, including a pair of claws and a pulvillus. Legs with all setae aciculate and smooth, most setae are relatively long, except leg III which has comparatively shorter setae, tarsi I–IV which have longest and thicker setae than leg segments especially tarsus IV and a set of very short setae as follows: *ad* in

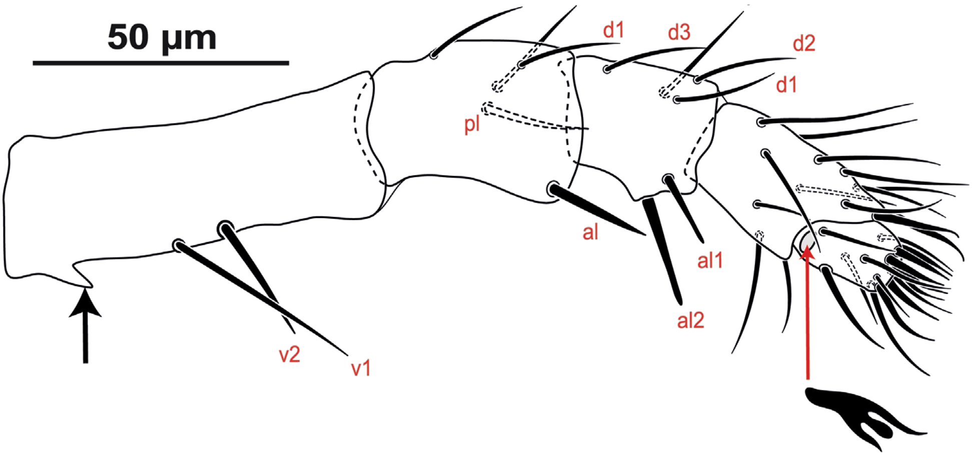

Figure 5. *Krantzolaspina angustatus* comb. nov., adult female. Palp, note the ventral spine-like process in palptrochanter.

trochanter I, *av* and ad_2 and al_2 in femur I, ad_{2-3} in femur II, al_{1-2} in genu II, *pl* and *pd* in femur IV and *pl* in genu IV.

Male. Unknown.

Material examined and depository. • 1♀ Holotype at NSMT (NSMT-Ac 9805) collected in Philippines, Palawan Islands, Olanguan Valley, between Puerto Princesa and Roxas, on litter of tropical forest. •16♀ vouchers at ESALQ collected in Iran, Kerman Province on soil and litter at the base of *Medicago sativa* (Fabaceae) and *Citrus sinensis* (Rutaceae). New material from Indonesia, Sumatra, Jambi Province, Bukit Duabelas region, upper soil layer (0–3 cm) of oil palm plantation, research site BO2b, 2°04'32.0"S, 102°47'30.7"E, 83.74 m a.s.l. 6♀ on slides as follows: • 1♀ deposited at LIPI (OSAL 00124840); • 1♀ deposited at SMNG (OSAL 00124839 SMNG 2020/62099). • 2♀ deposited at OSAL (OSAL 00124841, 00124842). • 2♀ deposited at ANIC (OSAL 00124843, 00124844). 10♀ in alcohol, as follows: • 2♀ deposited at LIPI, • 3♀ deposited at SMNG, • 2♀ deposited at OSAL and • 3♀ deposited at ANIC. All specimens were collected in November 2013 by B. Klarner. Additional photos of the species are deposited in Ecotaxonomy database at www.ecotaxonomy.org (ECOTAX_ID: 434549).

Differential diagnosis. *K. angustatus* comb. nov. significantly differs from *K. rebatii* and *K. solimani* comb. nov. in the following combination of characters: *K. angustatus* has 32 pairs of smooth dorsal setae (except *S5* and *Z5* slightly pilose), while *K. rebatii* and *K. solimani* have 36 pairs of smooth setae, respectively; *K. angustatus* has 15 pairs of setae *r-R* and *UR*-series in the unsclerotised lateral cuticle, whereas *K. solimani* and *K. rebatii* have 13 and 10, respectively; in *K. angustatus* and *K. solimani,* the peritreme is extended anteriorly between coxae I–II at the level of *st1*, while in *K. rebatii,* it is extended beyond coxa I; in *K. angustatus,* a spine-like process is developed in the subcapitulum, while this is absent in *K. rebatii* and *K. solimani*. Further, the

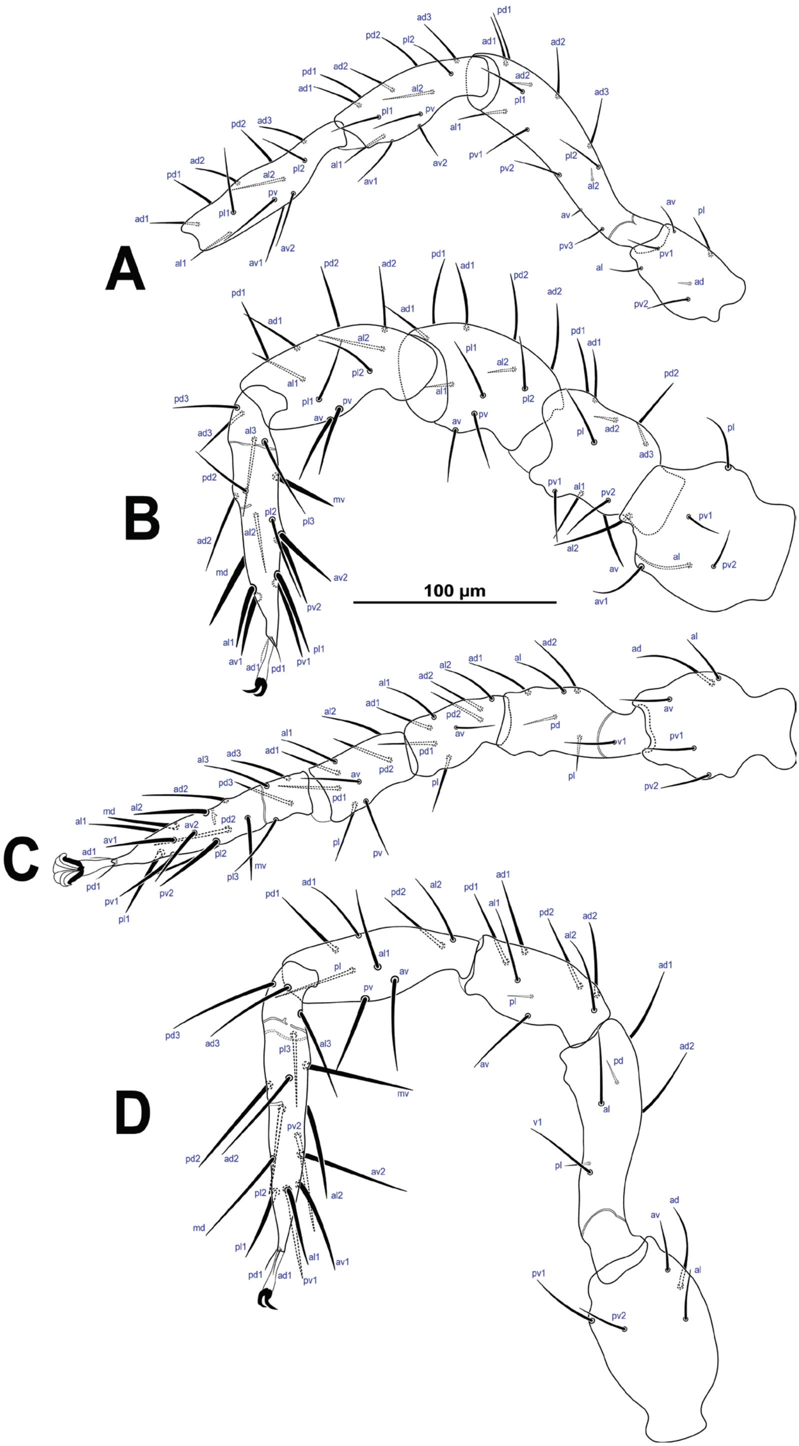

Figure 6. *Krantzolaspina angustatus* comb. nov., adult female. **A–D** Legs I–IV, respectively. Coxae I–IV in the Fig. 2.

Table 1. Characteristics of the females of *Krantzolaspina angustatus* comb. nov., and some ambiguous or unknown data of *K. rebatii* and *K. solimani* comb. nov.

Characters	***K. angustatus*** **(Ishikawa, 1987) comb. nov.**	******K. rebatii*** **Datta & Bhattacharjee, 1989**	******K. solimani*** **(Metwali, 1983) comb. nov.**
Dorsal shield ornamentation	mostly reticulate (smooth on the dorso-central region around bases of *z1* and *J2*)	completely smooth?	with tetra- and pentagonal reticulation (except on dorsocentral region of setae *j–J*, faintly ornamented)
Sternometasternal / ventrianal shield ornamentation	completely reticulate and with the lateral margins covered by some semi-rounded cells	mostly smooth but with lateral margins pointed?/ anterior margin pointed, elsewhere smooth?	mostly with tetra and pentagonal reticulation
Parapodal-peritremal shield ornamentation	slightly reticulate + semi-rounded cells posterior to the coxa IV	completely smooth?	completely smooth?
[1]Setae of opisthogastric cuticle (*JV–ZV*)	Seven pairs (*JV3–5*, *ZV2–5*); *JV4–5* and *ZV4–5* pilose	Six pairs (*JV4–5*, *ZV2–5*)?	Six pairs (*JV4–5*, *ZV2–5*)
Deutosternum: no. rows	Nine	three?	?
Leg chetotaxy (Coxa – Tibia)			?
I	2, 6, 13, 12, 12, not counted	?, ?, 10, 11, 11, ?	
II	2, 5, 11, 10, 10, 18	?, 4, 10, 9, ?, ?	
III	2, 5, 6, 8, 8, 18	?, 3, 5, 8, 7, ?	
IV	1, 5, 6, 8, 8, 18	?, ?, 4, 6, 7, ?	

? indicates unknown or uncertain/unclear data. [1]All setae are smooth and moderately long, except when mentioned otherwise. * holotype not found, presumed lost; characters presented are based on the original description.

arthrodial process in *K. rebatii* has a pair of long unequal arthrodial brushes, whereas the arthrodial process in *K. solimani* and *K. angustatus* has a relatively long arthrodial brush and a narrow fringed arthrodial corona. Additionally, in *K. angustatus*, small metapodal platelets elliptical in shape are present, while in *K. solimani* and *K. rebatii*, they are absent. Lastly, Table 1 provides uncertain/unclear or unknown characters of these species.

Remarks. *Krantzolaspina* is a well-defined genus by the unique combination of characters stated above (see diagnosis of the genus). However, a number of characters are also present in other dermanyssine families, for example, well-developed arthrodial brush(es) is/are also present in macrochelid mites, a sternometasternal shield bearing *st1–st4* is shared with species of Pachylaelapidae (Mašán and Halliday 2014) and Ologamasidae (Castilho et al. 2016) and the fusion of genital + ventral + anal shields forming a genitiventrianal shield is shared with two genera of Laelapidae (*Ololaelaps* and *Oloopticus*) (Beaulieu et al. 2019).

Although the genitiventrianal shield is a diagnostic character for *Krantzolaspina*, it is not an exclusive character as it also occurs in other genera of Parholaspididae, such as *Holaspulus*, some species of *Holaspina* and *Proparholaspulus*, as well as in other families, such as Leptolaelapidae genus *Indutolaelaps* Karg, 1997. In addition, this feature occurs in some species of Laelapidae (see above), but in those species, the genitiventrianal shield is hyper-developed posteriorly and occupies most of the opisthogaster and is often named the hologastric shield (Beaulieu et al. 2019).

One particular feature of *Krantzolaspina* is the pointed spur-like process present on the palp trochanter and this character is important for recognising the genus. However, a similar process is present in species (and all post-embryonic stages) of the monotypic family Megalolaelapidae (*Megalolaelaps*), in which the palp trochanter typically bears a large anteroventral horn-like projection. Unfortunately, the function of these processes of the palp trochanter in *Krantzolaspina* and *Megalolaelaps* is unknown (Mašán and Halliday 2014, Cómbita-Heredia et al. 2018). It should be noted that members of Megalolaelapidae are more similar morphologically to Pachylaelapidae and to Macrochelidae of the genus *Neopodocinum* Oudemans (Macrochelidae) than to Parholaspididae (Cómbita-Heredia et al. 2018).

Distribution of *Krantzolaspina* spp.

Krantzolaspina species have been collected from soils, mainly disturbed soil and in countries of Southern Asia. However, its distribution is isolated since they are present in Iran, Philippines and recently recorded in Indonesia (see details of locality type and other records, Table 1). A similar pattern of isolated distribution is present in other species of the parholaspidid family (e.g. *Holaspina alstoni* and *Parholaspis kewensis*, *P. meridionalis*) which were collected in botanical gardens from England where these records were based on accidental introduction (Ishikawa 1980c, Latifi et al. 2006) and potentially facilitated by human activity (Latifi et al. 2006). Hypothetically, we believe that these records of *Krantzolaspina* are based on introduced specimens through the trading of vegetal material including soil where they inhabit.

Notes on Indutolaelaps jiroftensis Hajizadeh et al., 2017 syn. nov. of *K. angustatus* (Ishikawa, 1987) comb. nov.

This species was described in the genus *Indutolaelaps* Karg, 1997 (Leptolaelapidae), based on a genitiventrianal shield, an epistome with an anteriomedial extension, wider at the base and acute distally and a palp tarsal claw three-tined (Hajizadeh et al. 2017). The misplacement of *I. jiroftensis* in the genus *Indutolaelaps* may suggest some genus-level similarity of *Indutolaelaps* and *Krantzolaspina*. These two genera indeed share several conspicuous characters such as similar shape of the genitiventrianal shield, presence of two pairs of presternal platelets, sternometasternal shield bearing *st1–4*, parapodal shield well-developed and fused with peritrematal shield, epistome with a narrow median projection and lateral margins serrate and 3-tined palptarsal claw. However, both genera which belong to different families can be distinguished by the combination of characters given in Table 2, such as arthrodial process, number of presternal platelets and preanal and dorsal setae.

Further, the synonymy of *I. jiroftensis* is supported in that it has 32 pairs of dorsal setae, two pairs of presternal platelets, sternometasternal and genitiventrianal shield with four pairs, as well a distinct ornamentation pattern (Hajizadeh et al. 2017 pp 670–671), which are diagnostic characters of the genus *Krantzolaspina* and specifically

Table 2. Morphological characteristics of females in the genera *Krantzolaspina* and *Indutolaelaps*.

Morphological characters	**Krantzolaspina* Datta & Bhattacharjee, 1989 (Parholaspididae)	***Indutolaelaps* Karg, 1997 (Leptolaelapidae)
Arthrodial process	with two arthrodial brush or one arthrodial brush and one narrow fringed arthrodial corona	one fringed arthrodial corona
Presternal platelelets	two pairs	one pair
Number of preanal setae in the genitiventrianal shield	three pairs	five pairs
Number of dorsal setae	32–34 pairs	50–55 pairs
Gnathotectum	with a median projection slightly bifurcate, trifurcate or serrate distally and with lateral margins serrate	with a median projection, distally and lateral margins smooth
Deutosternal rows	six, nine	five
Cheliceral dentition	FD usually with four teeth (rarely three) and MD bidentate	FD with three teeth and MD bidentate
Pretarsi	pretarsus I reduced or absent, pretarsi II–IV well developed	Pretarsus I reduced, pretarsi II–IV well developed

The present differential characters are listed in order of importance. [1]All setae are smooth and moderately long, except when mentioned otherwise.
* based on the present review; ** based on the original description of Karg (1997). FD – fixed digit; MD – mobile digit.

of *K. angustatus*. Additionally, the characters present in their drawings (figs 3, 5–7) and our photos (Figs 3B, 4B, E, H, K) of *I. jiroftensis* (Hajizadeh et al., 2017) match the characters from the holotype of *K. angustatus* (see Figs 3A, 4A, D, G, J).

Notes on *Krantzolaspina rebatii* Datta & Bhattacharjee, 1989

Krantzolaspina rebatii Datta & Bhattacharjee, 1989: 411

The holotype of *K. rebatii* was deposited according to Datta and Bhattacharjee (1989) in "Collection of Animal Ecology Laboratory, Department of Zoology, Gauhati University, Guwahati, India"; however, we were unable to locate this type specimen despite significant efforts. A careful study of type material will be essential to identify the diagnostic traits of that species.

Focusing on the original description, we like to mention some discrepancies and/or mistakes that we found between the text and illustrations as follows:

(1) *Dorsal shield*: "36 pairs of setae" is indicated in the text (Datta and Bhattacharjee 1989:411); however, their illustration (fig. 1b) shows only 34 apparent pairs of setae, although five represented only by sockets (presumably because the setae had fallen off). In addition, it is unclear whether the dorsal shield is smooth or not, as any type of ornamentation seems to be excluded from their original drawings and text.

(2) *Venter*: The ornamentation seems to be excluded from their original text, but it looks mostly smooth in fig. 1a of Datta and Bhattacharjee (1989) as well the lateral region punctate of the sternometasternal shield and the margin anterior of the genitiventrianal shield.

(3) *Legs*: Datta and Bhattacharjee (1989) provided an illustration of the legs (Fig. 1i–h), but without accompanying text in the description. The illustrations indicate a reduced number of setae compared with the leg chaetotaxy herein described for *K. angustatus* (Table 1). We presume that some setae were overlooked and not drawn by Datta and Bhattacharjee (1989).

Notes on *K. solimani* (Metwali, 1983) comb. nov.

Neoparholaspulus solimani Metwali, 1983: 459.

Metwali (1983) placed this species in the genus *Neoparholaspulus*, based on some characters that are typical for the genus, such as a genitiventrianal shield and one pair of presternal platelets, as well as one pair of metasternal shields free. In addition, Metwali's description of the species includes "metasternal plate well developed and free". However, the description also states that the sternal shield has four pairs of setae and the illustration shows that the metasternal plates are fused to the sternal shield. We have provisionally placed this species in *Krantzolaspina*, based on the presence of 32 pairs of setae in the dorsal shield, two pairs of presternal platelets and the assumption that fig. 2 of Metwali (1983) is inaccurate and that the metasternal plates are indeed fused to the sternal shield. Unfortunately, as the type specimens are lost, this interpretation cannot be confirmed (Reham Abo-Shnaf, personal communication).

***Krantzolaspina* sp.?**

Nawar and El-Sherif (1995: 273) re-described the female of a species that they identified as *Holaspina solimani* and described the male for the first time. Hussein et al. (2002: 1117) reared this species in the laboratory and studied its biology and behaviour. However, the illustrations and description in Nawar and El-Sherif (1995) differ in the grade of fusion of the metasternal plate, as well as the number of setae in the sternal shield from those in Metwali (1983) and we provisionally assume that these two specimens are two different species. Unfortunately, the specimens examined by Nawar and El-Sherif (1995) and Hussein et al. (2002) are lost and this interpretation cannot be confirmed (Reham Abo-Shnaf, personal communication).

In conclusion, the genus *Krantzolaspina* currently includes three valid species *Krantzolaspina angustatus* (Ishikawa, 1987) comb. nov., *K. rebatii* Datta & Bhattacharjee, 1989 and *K. solimani* (Metwali, 1983) comb. nov. Lastly, despite the valuable work undertaken by Krantz (1960), Petrova (1967, 1970, 1977), Ishikawa (1980a, 1980b, 1987b), Datta and Bhattacharjee (1989, 1991), Lee and Lee (2000) and the more recent works by Bhattacharyya and Kheto (2016) for *Proparholaspulus* and by Marchenko (2016) for *Neparholaspis*, it is clear that a revision of the family Parholaspididae is needed to know the current status.

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Parahiraciini (Hemiptera, Fulgoromorpha, Issidae): one new genus, two new species and three new subtribes

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Abstract

A new genus *Pusulissus* **gen. nov.** with two new species *P. phiaoacensis* **sp. nov.** and *P. coronomensis* **sp. nov.** are described respectively from Vietnam and China in the tribe Parahiraciini of the family Issidae. A molecular phylogeny using combined genes (18S, 28S, COX1 and Cytb) confirms its placement in the tribe Parahiraciini. The tribe is divided into three subtribes: Scantiniina **subtribe nov.** with the type genus *Scantinius* Stål, 1866, and Vindilisina **subtribe nov.** with type genus *Vindilis* Stål, 1870 plus *Nisoprincessa* Gnezdilov, 2017, and the nominal subtribe Parahiraciina Cheng & Yang, 1991 **subtribe nov.** The characteristics of these subtribes are provided, with a key to identification. Genus *Folifemurum* Che, Zhang & Wang, 2013 is transferred to Hemisphaeriini Mongolianina, and genus *Gelastyrella* Yang, 1994 is maintained as a valid name.

Keywords

Auchenorrhyncha, Fulgoroidea, molecular, morphology, phylogeny, planthopper, taxonomy

Introduction

The classification of the planthopper family Issidae Spinola, 1839 has been greatly modified in the recent years (reviewed in Wang et al. 2016). Emeljanov (1990) recognized seven subfamilies in the family Issidae Spinola, 1839 (including Acanaloniinae Amyot & Serville, 1843, Bladininae Kirkaldy, 1907, Caliscelinae Amyot & Serville, 1843, Tonginae Kirkaldy, 1907 and Trienopinae Fennah, 1954 which have now been dispatched to other planthopper families). In comparison Cheng and Yang (1991a, b) restricted the family Issidae to only five subfamilies: Issinae, Tonginae, Caliscelinae, Hemisphaeriinae Melichar, 1906 and an additional new monogeneric subfamily Parahiraciinae Cheng & Yang, 1991. This last taxon was based on the genus *Parahiracia* Ôuchi, 1940, originally described within the family Tropiduchidae before being transferred into Issidae by Fennah (1982). This distinction was not followed by Chan and Yang (1994: 86) and Emeljanov (1999) who transferred the genus to the issid Thioniini Melichar, 1906. Only ten years later, Gnezdilov (2003: 306) recognized again *Parahiracia*'s particular taxonomic position in a separate monospecific tribe Parahiraciini Cheng & Yang, 1991, based on its highly modified hindwing. This separation was confirmed later by molecular analyses (Wang et al. 2016).

A year after Parahiraciini's rehabilitation, Gnezdilov et al. (2004: 221) synonymized *Parahiracia* under *Fortunia* Distant, 1909 and added *Scantinius* Stål, 1866, *Pterygoma* Melichar, 1903, *Prosonoma* Melichar, 1906 (synonymized later with *Bardunia* Stål, 1863) and *Bardunia* to this tribe. In addition to the long incision between the first two hindwing lobes, the authors characterized the tribe by 'a strongly protruding metope in shape of probocis and well developed bi- or tri-lobed hindwings'. Another year later, Gnezdilov and Wilson (2005) added two more genera: *Pinocchias* Gnezdilov & Wilson, 2005, *Narinosus* Gnezdilov & Wilson, 2005 and synonymized *Clipeopsilus* Jacobi, 1944 with *Fortunia*. In 2007, Gnezdilov and Wilson transferred *Pterygoma* to Caliscelidae, but added *Mincopius* Distant, 1909 and *Flavina* Stål, 1861 to the tribe, with a key to the seven Parahiraciini genera (Gnezdilov and Wilson 2007). Zhang and Chen (2008, 2009, 2010, 2012) successively added five more genera: *Neodurium* Fennah, 1956, *Tetricodes* Fennah, 1956, *Fusiissus* Zhang & Chen, 2010, *Paratetricodes* Zhang & Chen, 2010, and *Neotetricodes* Zhang & Chen, 2012. In 2013, *Folifemurum* Che, Zhang & Wang, 2013 was described (Che et al. 2013). In his review of Issidae of the world, Gnezdilov (2013) added *Duriopsilla* Fennah, 1956, listing therefore 14 genera in the tribe.

Chen et al. (2014) re-established *Gelastyrella* Yang, 1994 as a valid taxon and one year later, Wang et al. (2015) and Meng et al. (2015) added respectively *Tetricodissus* Wang, Bourgoin & Zhang, 2015 and *Brevicopius* Meng, Qin & Wang, 2015 to the Parahiraciini. In the same year, Gnezdilov (2015) discussed the possibility that *Pseudochoutagus* Che, Zhang & Wang, 2011 and *Thabena* Stål, 1861 (including its synonyms *Cibyra* Stål, 1861, *Gelastyra* Kirkaldy, 1904, *Gelastyrella* Yang, 1994 (sic: not mentioning Chen et al. (2014)'s rehabilitation) and *Borbonissus* Bonfils, Attié & Reynaud, 2001) might belong to Parahiraciini. The transfer of these two genera as well as *Macrodarumoides* Che, Zhang & Wang, 2012, was formally confirmed by Wang et al. (2016), supported by molecular phylogeny analyses. However, they excluded *Folifemurum* moving it to Hemisphaeriini (Hemisphaeriina). The authors briefly characterized further Parahiraciini with the hindwing conformation: a deep narrowed incision of hindwing with a Pcu-A1 lobe distinctly wider than ScP-R-MP-Cu lobe, a short and thin A2 lobe in which A2 vein is often absent, free Pcu and A1, not partially fused. The same year, *Rhombissus* Gnezdilov & Hayashi, 2016 was described (Gnezdilov and Hayashi 2016). Finally, Gnezdilov (2017) added *Nisoprincessa* Gnezdilov, 2017 and *Thabenula* Gnezdilov, Soulier-Perkins & Bourgoin, 2011, and transferred the Philippines genus *Vindilis* Stål, 1870 into the tribe (Gnezdilov 2018).

Interestingly, Parahiraciini are also known from one fossil genus, *Bolbossus*† Gnezdilov & Bourgoin, 2016, from Baltic amber (dated from Priabonian: 37.8–33.9 Mya) (Gnezdilov and Bourgoin 2016).

In fact, in some 15 years since Parahiraciini was recognized again by Gnezdilov (2003), the tribe shows already a rather complex history with up to 26 genera with 82 species (Bourgoin 2020). In this study we retain 26 genera including the new one described. Figure 1 summarizes this already rich and complex chronological account of Parahiraciini generic taxa.

In addition to this synthetic taxonomic review, we also describe a new genus (already mentioned in Wang et al. (2016)'s molecular phylogeny under the name 'Gen. nov. *apud Tetricodes*') with two new species from Vietnam and China. A molecular phylogeny, using the combined genes (18S, 28S, COX1 and Cytb) of the already sequenced taxa, allows us to place this new genus into Parahiraciini and to briefly discuss this placement. Moreover, the large diversity of the taxa now included in the tribe also allows us to better clarify its morphological characteristics. Accordingly, the tribe is divided into three subtribes: Scantiniinia subtribe nov., Vindilisina subtribe nov. and the nominal subtribe Parahiraciina Cheng & Yang, 1991 subtribe nov., for which morphological characteristics are compared.

Materials and methods

Morphological interpretations and subsequent terminologies follow Bourgoin (1987) and Bourgoin (1993) respectively for male and female genitalia and Bourgoin et al. (2015) for wing venation. The metatibiotarsal formula provides the number of spines on the side of the metatibia – apex of metatibia/apex of first metatarsomere/apex of second metatarsomere. Issidae classification follows Wang et al. (2016)'s phylogeny, adapted in Bourgoin et al. (2020).

All the type specimens of the species described below are deposited in the Museum national d'Histoire naturelle (MNHN), Paris, France or in the China West Normal University (CWNU), Nanchong, Sichuan Province, China.

For genitalia study, the abdomen was separated from the specimen using microscissors, and then boiled in a 10% NaOH solution for several minutes until the muscles were completely dissolved leaving only tegumentary structures. Final dissection and observation of the abdomen was done in glycerin after rinsing in distilled water

			Parahiracia Ôuchi, 1940 [Tropiduchidae]	Ôuchi (1940)
			Parahiracia Ôuchi, 1940 [to Issidae]	Fennah (1982)
Issidae	Parahiraciinae Cheng & Yang, 1991	←	*Parahiracia* Ôuchi, 1940	Cheng and Yang (1991a)
Issidae Thioniini	X	→	*Parahiracia* Ôuchi, 1940 [to Issidae Thioniini]	Chan and Yang (1994)
		/	*Dindinga* Distant, 1909 [syn. of *Scantinius* Stål, 1866]	Liang (2001)
Issidae Hemisphaeriinae	Parahiraciini Cheng & Yang, 1991	←	*Parahiracia* Ôuchi, 1940 [from Issidae Thioniini]	Gnezdilov (2003)
		←	*Prosonoma* Melichar, 1906	"
		←	*Fortunia* Distant, 1909	Gnezdilov et al. (2004)
		→	*Parahiracia* Ôuchi, 1940 [syn. of *Fortunia* Distant, 1909]	"
		←	*Scantinius* Stål, 1866 - [to Scantiniina **subtrib.nov.**, this study]	"
		←	*Pterygoma* Melichar, 1903	"
		←	*Bardunia* Stål, 1863	"
		←	*Pinocchias* Gnezdilov & Wilson, 2005	Gnezdilov and Wilson (2005)
		←	*Narinosus* Gnezdilov & Wilson, 2005	"
		/	*Clipeopsilus* Jacobi, 1944 [syn. of *Fortunia* Distant, 1909]	"
		→	*Pterygoma* Melichar, 1903 [to Caliscelidae]	Gnezdilov and Wilson (2006)
		→	*Prosonoma* Melichar, 1906 [syn. of *Bardunia* Stål, 1863]	Gnezdilov and Wilson (2007)
		←	*Flavina* Stål, 1861	"
		/	*Nilalohita* Distant, 1906 [syn. of *Flavina* Stål, 1861]	"
		←	*Mincopius* Distant, 1909	"
		←	*Neodurium* Fennah, 1956	Zhang and Chen (2008)
		←	*Tetricodes* Fennah, 1956	Zhang and Chen (2009)
		/	*Cibyra* Stål, 1861[*nom. praeoc.*]	Gnezdilov (2009)
		/	*Dolia* Kirkaldy, 1907 [syn. of *Flavina* Stål, 1861]	"
		/	*Gelastyra* Kirkaldy, 1904 [syn. of *Thabena* Stål, 1866]	"
		/	*Gelastyrella* Yang, 1994 [syn. of *Thabena* Stål, 1866]	"
		/	*Borbonissus* Bonfils, Attié & Reynaud, 2001 [syn. of *Thabena* Stål, 1866]	"
		←	*Fusiissus* Zhang & Chen, 2010	Zhang and Chen (2010)
		←	*Paratetricodes* Zhang & Chen, 2010	"
		←	*Neotetricodes* Zhang & Chen, 2012	Zhang and Chen (2012)
		←	*Folifemurum* Che, Zhang & Wang, 2013	Che et al. (2013)
		←	*Duriopsilla* Fennah, 1956	Gnezdilov (2013)
		←	*Gelastyrella* Yang, 1994 [nom. rest.]	Zhang and Chang (2014)
		←	*Tetricodissus* Wang, Bourgoin & Zhang, 2015	Wang et al. (2015)
		←	*Brevicopius* Meng, Qin & Wang, 2015	Meng et al. (2015)
		←	*Rhombissus* Gnezdilov & Hayashi, 2016	Gnezdilov and Hayashi (2016)
		←	*Thabena* Stål, 1861	Wang et al. (2016)
		←	*Pseudochoutagus* Che, Zhang & Wang, 2011	"
		←	*Macrodarumoides* Che, Zhang & Wang, 2012	"
		→	*Folifemurum* Che, Zhang & Wang, 2013 [to Hemisphaeriina]	"
		←	*Bolbossus*† Gnezdilov & Bourgoin, 2016	Gnezdilov and Bourgoin (2016)
		←	*Nisoprincessa* Gnezdilov, 2017 - [to Vindilisina **subtrib.nov.**, this study]	Gnezdilov (2017b)
		←	*Folifemurum* Che, Zhang & Wang, 2013	"
		←	*Thabenula* Gnezdilov, Soulier-Perkins & Bourgoin, 2011	"
		←	*Vindilis* Stål, 1870 - [to Vindilisina **subtrib.nov.**, this study]	Gnezdilov (2018)
		←	*Duroides* Melichar, 1906	Gnezdilov (2019)
		←	*Pusulissus* Bourgoin & Wang, 2020	This study
		→	*Folifemurum* Che, Zhang & Wang, 2013 [to Mongolianina]	"

Figure 1. Chronological account of the genera in the tribe Parahiraciini Cheng & Yang, 1991. Box colors follows FLOW (Bourgoin 2020) standard colors with pale blue: taxon protonym and green: accepted taxon. Left arrows indicate when a genus was included in the tribe and right ones when excluded. Red and blue arrows refer respectively to taxa excluded or included in Parahiraciini.

several times. Genitalia were finally stored in genitalia vials under the corresponding specimen. Photographs of external morphology and genitalia characters were taken using a Leica DFC camera attached to a Leica M205FA stereomicroscope and further refined with the software LAS X.

Total genomic DNA was extracted from the fore or middle leg from a paratype of *Pusulissus phiaoacensis* sp. nov. and the holotype of *Pusulissus coronomensis* sp. nov. using a Sangon Ezup column animal genomic DNA purification kit. The DNA of the genes (18S rRNA, 28S rRNA, COXI, Cytb) was amplified using the same primers and amplification procedures as in Wang et al. (2016). DNA sequencing was conducted by the Sangon Company (Shanghai, China). Contigs assembly was made using the software Seqman from package DNAstar v5.01 (www.dnastar.com). All sequences obtained in this study were registered in GenBank with accession numbers mentioned below.

MEGA v7.0 (Kumar et al. 2016) was used for performing alignments for a subset of Parahiraciini taxa already analysed in Wang et al. (2016) plus the specimen of *Rhombissus* sp. analysed in Zhao et al. (2019). Four species (two species in Sarimini and two species in Hemisphaeriini) were chosen as outgroups for the analysis. The related taxon name, collecting location, and GenBank accession numbers are in Table 1. Phylogenetic analysis was performed using the software MrBayes v.3.2.4 (Ronquist et al. 2012) using the same method as Wang et al. (2016) except for running with 100 millions generations, sampling every 1000 generations. FIGTREE v1.1.2 (Rambaut 2016) was used to visualize the tree.

Taxonomy

Family Issidae Spinola, 1839
Subfamily Hemisphaeriinae Melichar, 1906 (sec. Wang et al. 2016)
Tribe Parahiraciini Cheng & Yang, 1991

Pusulissus gen. nov.

http://zoobank.org/CBD98E0A-F175-45B4-8199-D57E7B1243A4
Figs 2–44

Type species. *Pusulissus phiaoacensis* sp. nov.

Diagnosis. This genus is similar to *Tetricodes* Fennah, 1956, from which it differs as follows: 1) The absence of the frontal black median tubercle on the disc of frons (Figs 5, 28); 2) Median carina of frons well distinct, extending from the dorsal margin almost to the frontoclypeal suture (Figs 5, 28), while it is only present in the dorsal part of the frons or invisible in *Tetricodes* (Zhang and Chen 2009, fig. 21); 3) The forewing distinctly broadest in the basal 1/3 (Figs 6, 29), while regularly convex in *Tetricodes* (Zhang and Chen 2009, fig. 4).

This genus is also very similar to *Thabena* Stål, 1866, but differs by 1) Its vertex with anterior margin in dorsal view very slightly angularly convex (Fig. 4) or straight (Fig. 27), while roundly convex in *Thabena* (Chen et al. 2014, figs 2–74C); 2) Frons much longer, more than 1.2 times longer in midline than widest part (Figs 5, 28), but wider than long in *Thabena*, less than 0.9 times longer in midline than widest part (Chen et al. 2014, fig. 2–74E).

Description. Head with the compound eyes a little wider than pronotum (Figs 4, 27). Vertex nearly rectangular, slightly wider than long, without median carina or median carina very weakly present; anterior margin very slightly protruded (Fig. 4) or almost straight (Fig. 27), lateral margins parallel, posterior margin angularly concave medially (Figs 4, 27). Frons obviously longer than wide, apical and lateral margins carinated and elevated, dorsal margin deeply concave at middle, frons slightly narrower in the middle level of the compound eyes, then gradually broadened

Table 1. Taxa sampling, collecting locations and GenBank accession numbers used for the phylogenetic study.

Species name	Collecting location	Gene 18S	Gene 28S (D3–D5)	Gene 28S (D6–D7)	COXI	Cytb
Fortunia sp.	China, Yunnan, Xishuangbanna, Mengla, Menglun, 21°24'398"N, 101°16'754"E, 705±21 m	KX761487	KX761527	KX761518	KX761498	KX761509
Flavina hainana (Wang & Wang, 1999)	China, Hainan, Jianfengling	KX702824	KX761453	MN381846		KX702912
Gelastyrella litaoensis Yang, 1994	China, Hainan, Bawangling	KX702823	KX761452	KX702811	KX761461	KX702911
Macrodarumoides petalinus Che, Zhang & Wang, 2012	China, Guangxi, Baise, Jinzhongshan, Songshuping	KX702827		KX702856	KX702926	KX702880
Neodurium hamatum Wang & Wang, 2011	China, Yunnan, Xishuangbanna, Mengla, Menglun, 21°24'398"N, 101°16'754"E, 705±21 m	KX702818	KX761446	MN381844	KX702920	
Tetricodissus pandlineus Wang, Bourgoin & Zhang, 2015	China, Yunnan, Xishuangbanna, Mengla, Nanshahe, 21°36'12.1"N, 101°34'23.9"E, 826±43 m	KX702817	KX761445	KX702807		KX702907
Tetricodes songae Zhang & Chen, 2009	China, Guizhou, Kuankuoshui, 1500 m	KX702841	KX761457		KX702925	KX702916
Pusulissus sp.	Vietnam, Vinh Phuc, Tam Dao, 21°26'47"N, 105°38'38"E, 748 m	KX761479		KX761485	KX761471	KX761475
Pusulissus coronomensis sp. nov.	China, Guangxi, Hezhou, Qichong, 24°13'6"N, 110°48'34"E, 180 m	MT772139	MT772137	MT772136	MT774094	MT774093
Pusulissus phiaoacensis sp. nov.	Vietnam, Phia Oac, 1050 m	MW201961			MT774095	MT774092
Rhombissus sp.	China, Shaanxi, Zhouzhi, Houzhenzi, 1050 m	MN381855	MN381852	MN381850		MN332231
Duplexissus punctatulus Wang, Zhang & Bourgoin, 2019	China, Yunnan, Xishuangbanna, Mengla, Menglun	KX761490	KX761531	KX761520	KX761501	KX761512
Hemisphaerius lysanias Fennah, 1978	Vietnam, Khanh Hoa Province, Hon-Ba massif, 12°13'20"N, 109°06'00"E	KX702833	KX761404	KX702860	KX702933	KX702883
Hemisphaerius coccinelloides (Burmeister, 1834)	Philippines, Los Banos, UP Hortarium, 14°09'53"N, 121°14'14"E	KX702834	KX761405	KX702861	KX702934	KX702884
Sarima bifurca Meng & Wang, 2016	China, Yunnan, Xishuangbanna, Mengla	KX702819	KX761447	KX702808	KX702921	KX761552

below the lower margin of the compound eyes (Figs 5, 28); median carina obviously elevated from the dorsal margin extending to near base, but not reaching to frontoclypeal suture (Figs 5, 28); disc with several tubercles in its dorsal part and lateral areas (Figs 5, 28). Frontoclypeal suture straight (Fig. 5) or slightly convex (Fig. 28). Clypeus flattened, without median carina or median carina very weak, almost invisible (Figs 5, 28). Rostrum long, reaching to hind coxae; third segment almost as long as second one. Genae in lateral view flattened and oblique, with a small protuberance near base (Figs 3, 26). Pronotum triangular, anterior and posterior margins elevated, without carina (Figs 4, 27), paranotal lobes developed. Mesonotum inverted triangular, a little longer than pronotum at midline, without carina (Figs 4, 27). Forewings twice longer than broad, longitudinal veins obvious and elevated, transverse veins not elevated, but existing in the whole forewing, apical margin oblique, triangular-shaped (Figs 6, 29). Vein ScP+R firstly separated near base after a short common stem, ScP+RA long, reaching apical 1/5 of costal margin, terminal of RP vein reaching to apical margin of forewing (Figs 6, 29); MP vein firstly forked near basal 1/4, MP_{1+2} forked again at apical 1/4, MP_{3+4} forked again at middle, MP_3 short, just reaching bifurcation of MP_1 and MP_2 or slightly beyond bifurcation, MP_4 long, extending to apical margin of forewing; CuA simple, sinuate, extending to apex of forewing (Figs 6, 29). Clavus closed, reaching to almost middle of forewing, Pcu and A1 fused at apical 1/3 of clavus (Figs 6, 29). Hindwing with longitudinal veins well developed, with a set of numerous transverse veins, CuP-Pcu-A1 lobe distinctly wider than ScP-R-MP-Cu lobe, A2 lobe very narrow with A2 vein absent (Figs 7, 30); MP and CuA not fused, Pcu and A1 also separated (Figs 7, 30). Hind tibia with 2 lateral spines on apical half (Fig. 3). Metatibiotarsal formula: 2-(7-8)/(5-8)/2.

Male genitalia. Anal tube in lateral view relatively large and thick, with lateral lobes on apical half extended downwards (Figs 8, 31). Pygofer subrectangular in lateral view (Figs 8, 31). Gonostyli nearly triangular in profile, dorsal margin sinuate, posterior margin deeply concave to a groove in apical half then strongly convex posteriorly, caudo-ventral angle strongly convex and rounded (Figs 10, 33). Capitulum broader than high, triangular, with a relatively long meniscate lateral process with both apical parts obtuse, apical process sharp (Figs 10, 33). Periandrium symmetrical, with dorsal and ventral margins parallel, shallowly U-shaped, divided into dorso-lateral lobe (dll) and ventral lobe (vl), ventral lobe very slightly shorter than dorso-lateral one (Figs 11, 13, 34, 36). Aedeagus (Ade) with a pair of lateral processes (Adep) originating from the middle, directed anteriorly and upcurved (Figs 11, 13, 34, 36).

Female genitalia. Gonoplacs in lateral view nearly rectangular, dorsal margin straight, anterior and posterior margins nearly parallel to each to other, posterior margin with apical half membranous, ventral margin with the needle-shaped basal part (Figs 16, 40); in dorsal view lateral margins convex outward, median area fused in apical half (Fig. 39). Gonapophysis IX in dorsal view widest a little beyond middle then gradually sharpening to apex (Figs 17, 41); in lateral view broad, widest near middle, divided into upper and lower parts, both with sharp tips, bifurcate near apex

(Figs 18, 42); upper dorsal margin sinuate, flattened at basal 1/3 and middle part obviously convex upward then gradually sloping posteriorly (Figs 18, 42); ventral lower margin with apical half sloping downwards (Figs 18, 42). Gonospiculum bridge small (Figs 17, 18, 41, 42). Anterior connective lamina of gonapophysis VIII with obscure teeth on apex and outer-lateral margin, inner-lateral margins without teeth (Figs 20, 44). Endogonocoxal process membranous, slightly shorter than anterior connective lamina of gonapophysis VIII (Figs 20, 44). Gonocoxa VIII connects with gonapophysis VIII by a rectangular shape (Figs 20, 44).

Etymology. The name is an arbitrary association from the Latin word "pusulosus" which means 'pustulous', referring to the tubercles or pustules present on the frons and "issus" referring to the family. The name is treated as masculine.

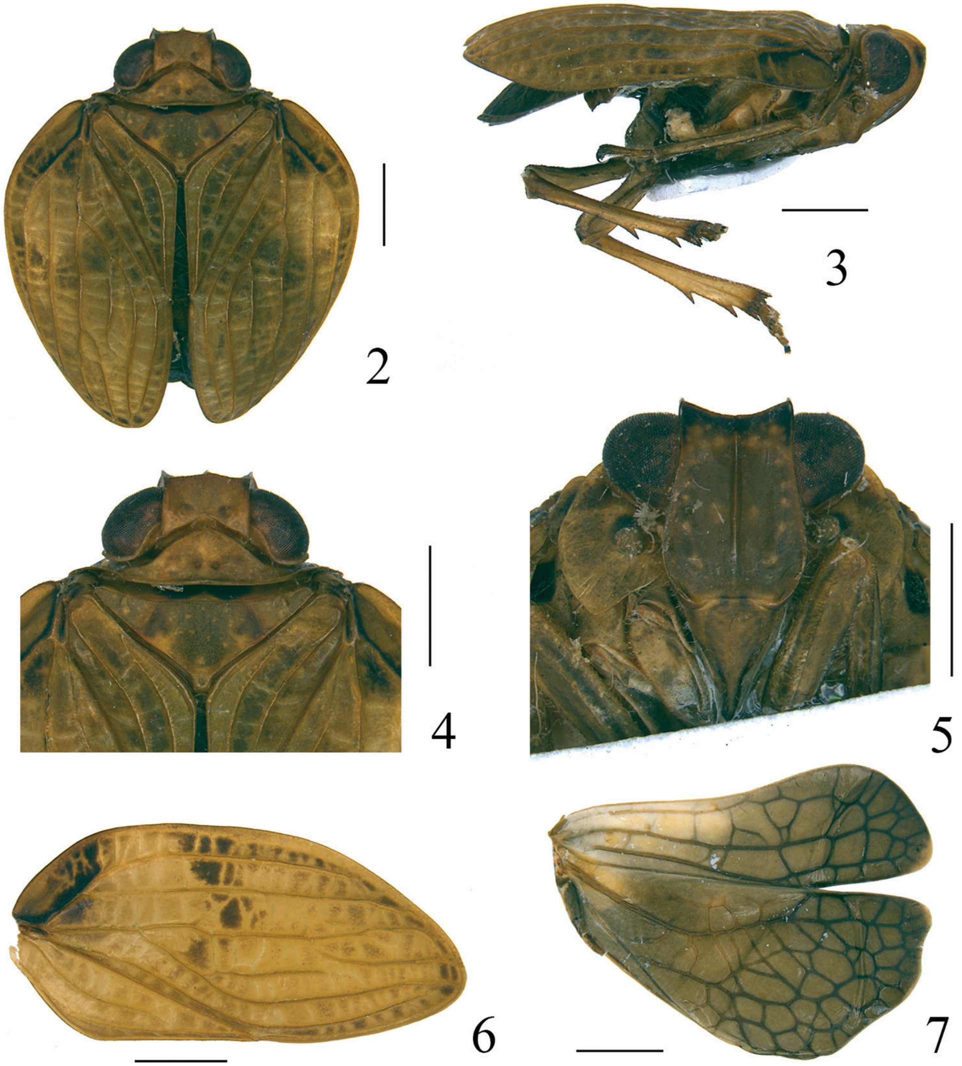
Pusulissus phiaoacensis **sp. nov.**

http://zoobank.org/8C827F6F-344C-419A-A1B1-CCFA648CF74B
Figs 2–24

Type materials. ***Holotype*:** ♂, VIETNAM: Phia Oac, 12 vii 2019, 1050 m, (22°26'0.78"N, 105°53'4.692"E), CAV [chasse à vue], rec. Th. Bourgoin, MNHN(EH) 24743. ***Paratypes*:** 1♀, same data as holotype, MNHN(EH) 24744 [sequenced specimen]; 1♀, same location and collection date as holotype, but rec. G. Kunz MNHN(EH) 24745.

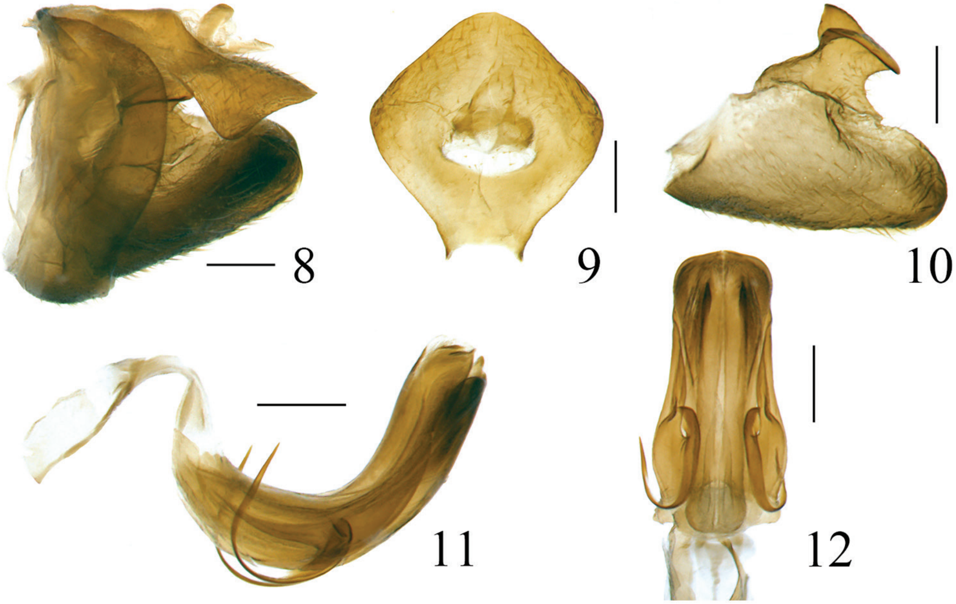
Description. Length: male (including forewings) (*N* = 1): 5.0 mm; female (including forewings) (*N* = 2): 5.1–5.3 mm.

Coloration. For the dry specimens, general coloration tawny (Fig. 2). Vertex tawny, with two brown circular markings near base of disc (Fig. 4). Compound eyes black, supported by tawny callus (Figs 4, 5). Frons brown, dorsal part black (Fig. 5); dorsal and lateral margins carinated and brown, median carina brown (Fig. 5). Frons with two tawny round markings in middle area, dorsal and lateral areas distributed with around ten tawny tubercles on each side (Fig. 5). Antennae brown (Fig. 5). Postclypeus brown (Fig. 5). Genae tawny (Fig. 3). Pronotum tawny, median area with two small brownish impressions on disc (Fig. 4), paranotal lobes tawny (Fig. 5). Mesonotum tawny, with two brown longitudinal bands on disc (Fig. 4). Forewings tawny with veins tawny, basal part of costal area with some irregular black markings, area from basal 1/3 of costal margin extending to middle of forewing surface also with some irregular black markings, apical margin with black linear marking (Fig. 6); some specimens with these black markings almost invisible, but with one extremely large whitish round marking at basal 1/3 of forewing on each side (Fig. 2). Hindwings brown, veins clearer (Fig. 7). Legs tawny (Figs 3, 5). For alive and fresh specimens, vertex, pronotum, mesonotum and forewings interspersed with some reddish markings or coloration as mentioned above (Figs 21–23). Compound eyes yellow (Figs 21–23). Frons brown, dorsal part black, median carina brown, disc distributed with several yellow tubercles (Fig. 22). Legs brown (Figs 21–23).

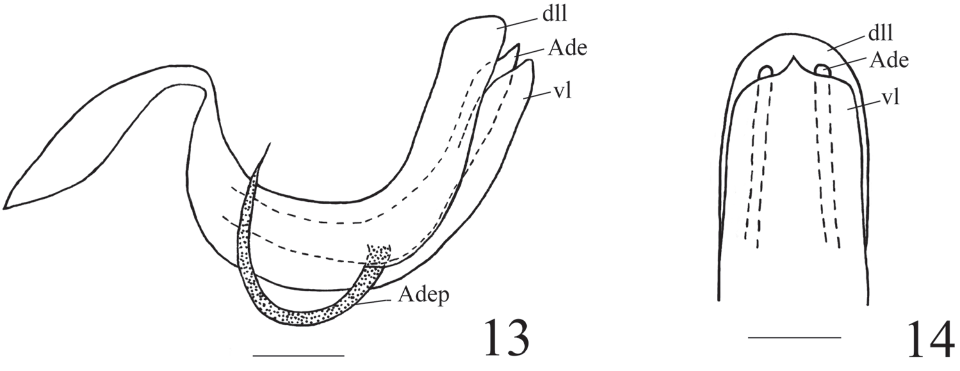


Figures 2–7. *Pusulissus phiaoacensis* sp. nov. **2** adult (holotype), dorsal view **3** adult (holotype), lateral view **4** head and thorax (holotype), dorsal view **5** adult (holotype), frontal view **6** forewing (paratype) **7** hindwing (paratype). Scale bars: 1 mm.

Head and thorax. Vertex 2.1 times wider than long in midline, anterior margin very slightly angularly convex (Fig. 4). Frons 1.3 times longer in midline than widest part, 1.3 times wider at widest part than dorsal margin (Fig. 5). Pronotum 1.4 times longer in midline than vertex in midline (Fig. 4). Mesonotum 2.3 times wider along anterior margin than long in midline (Fig. 4). Forewings 2.1 times longer at longest part than widest part (Fig. 6). Hindwing with apical margin of CuP-Pcu-A1 lobe sinuate (Fig. 7). Metatibiotarsal formula: 2–8/5/2.

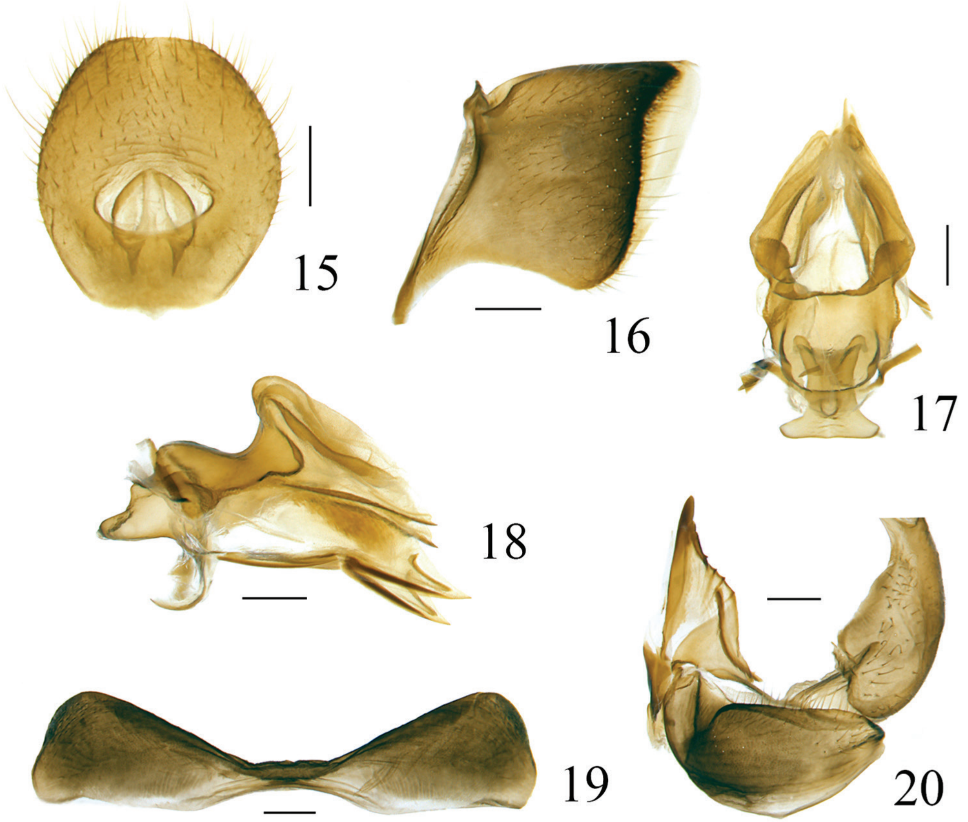


Figures 8–12. *Pusulissus phiaoacensis* sp. nov., male, holotype. **8** genitalia, lateral view **9** anal tube, dorsal view **10** gonostylus, lateral view **11** phallic complex, right lateral view **12** phallic complex, ventral view. Scale bars: 0.2 mm.



Figures 13–14. *Pusulissus phiaoacensis* sp. nov., male, holotype. **13** phallic complex, right lateral view **14** apex of phallic complex, posterior view. Abbreviations: dll: dorso-lateral lobe of periandrium; Ade: Aedeagus; vl: ventral lobe of periandrium; Adep: Aedeagus processes. Scale bars: 0.2 mm.

***Male genitalia*.** Anal tube in lateral view with lateral lobes well visible (Fig. 8); in dorsal view mushroom shaped, widest at middle, as long in midline as widest part, apical margin angularly rounded, lateral margins strongly angularly rounded (Fig. 9); anal opening located near middle of anal tube (Fig. 9). Pygofer in lateral view 2.5 times higher than wide, dorsal margin obviously sloping to the posterior, dorso-lateral angle rounded, posterior margin strongly roundly convex (Fig. 8). Gonostylus in lateral view with dorsal margin slightly sinuate (Fig. 10). Capitulum of gonostylus derived from



Figures 15–20. *Pusulissus phiaoacensis* sp. nov., female, paratype. **15** anal tube, dorsal view **16** gonoplac, lateral view **17** gonapophysis IX and gonospiculum bridge, dorsal view **18** gonapophysis IX and gonospiculum bridge, lateral view **19** sternite VII, ventral view **20** gonocoxa VIII and gonapophysis VIII, lateral view. Scale bars: 0.2 mm.

middle of gonostylus, broad, sharp triangular, directed to anterior; meniscate processes with one side not reaching to anterior margin of capitulum and another side beyond posterior margin of capitulum (Fig. 10). Periandrium in lateral view with dorso-lateral lobe rounded apically (Figs 11, 13). In posterior view, ventral lobe of periandrium with apical margin mostly straight but spinous protruded at middle (Fig. 14). Lateral processes of aedeagus long and slender, hooked, derived from the middle, directed anteriorly to basal 1/4, then curved upward, surpassing the dorsal margin of periandrium (Figs 11, 13); in ventral view this pair of processes curved outwards (Fig. 12).

Female genitalia. Anal tube in dorsal view broadly ovate, 1.1 times longer in midline than widest part, widest at middle, apical margin nearly straight, lateral margins rounded, anal opening situated slightly below middle (Fig. 15). Anterior connective lamina of gonapophysis VIII with two obscure large teeth in the apex and four small keeled teeth in outer-lateral group (Fig. 20). Gonocoxa VIII subquadrangular (Fig. 20). Hind margin of sternite VII roundly concave, with the median part nearly straight (Fig. 19).

Figures 21–24. Habitus and habitat of *Pusulissus phiaoacensis* sp. nov. **21–23** habitus in nature (Photo G. Kunz) **24** habitat (Photo T. Bourgoin).

Etymology. The name refers to the locality where the new species was found.

Habitat. The species was swept from pteridophytes at the margin of the mountainous rainforest (around 1050 m altitude) beside a road (Fig. 24).

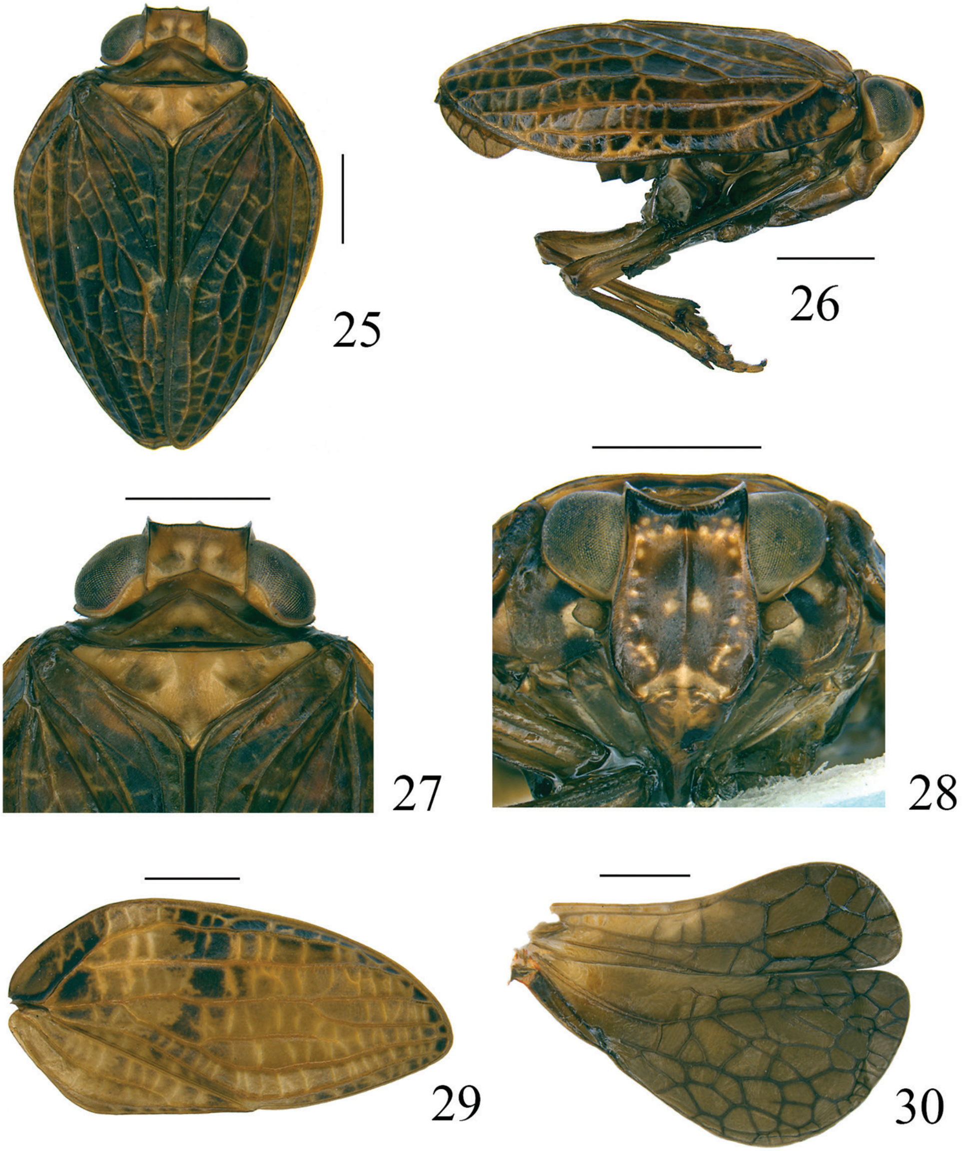
Note. Genes sequences were registered in GenBank with the following accession numbers: MW201961 (18S), MT774095 (COXI), MT774092 (Cytb). *P. phiaoacensis* differs from another unnamed *Pusulissus* species (referred here as *Pusulissus* sp.) (Fig.46) by 25 bp in the length 681 bp of COXI. Both species are from Vietnam.

Pusulissus coronomensis sp. nov.

http://zoobank.org/7537121D-C88F-44EC-8E7E-47696162D02A

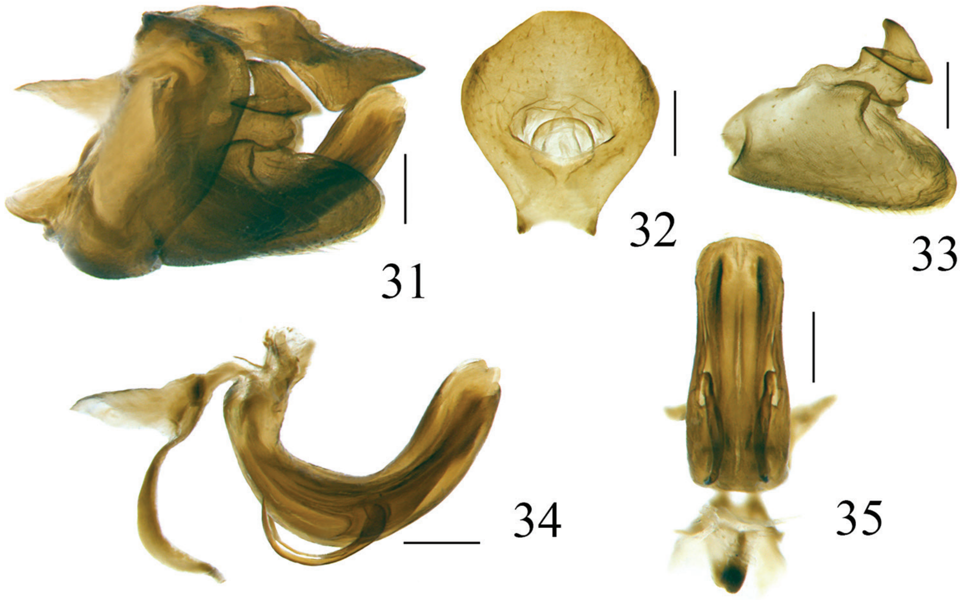
Figs 25–44

Type materials. ***Holotype*:** ♂, China: Guangxi Province, Hezhou, Qichong natural reserve, 24°13'6"N, 110°48'34"E, 180 m, 7 viii 2018, coll. Feilong Yang & Kun Zhao (CWNU) [sequenced specimen]. ***Paratypes*:** 1♂, 1♀, same data as holotype (CWNU).

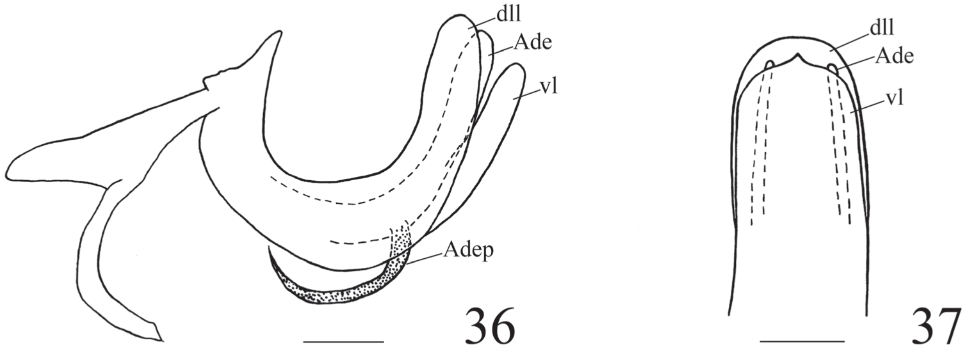


Figures 25–30. *Pusulissus coronomensis* sp. nov. **25** adult (holotype), dorsal view **26** adult (holotype), lateral view **27** head and thorax (holotype), dorsal view **28** adult (holotype), frontal view **29** forewing (paratype) **30** hindwing (paratype). Scale bars: 1 mm.

Differential diagnosis. This new species is very similar to *P. phiaoacensis* from Vietnam, but differs as follows: 1) Apical margin of the Pcu-A1 lobe on hindwing round (Fig. 30), while in *P. phiaoacensis* it is sinuate (Fig. 7); 2) Pair of lateral processes on aedeagus shorter, reaching to the ventral margin of periandrium (Fig. 34), while in *P. phiaoacensis* they surpass the dorsal margin of periandrium (Fig. 11); 3) Dorsal margin



Figures 31–35. *Pusulissus coronomensis* sp. nov., male, holotype. **31** genitalia, lateral view **32** anal tube, dorsal view **33** gonostylus, lateral view **34** phallic complex, right lateral view **35** phallic complex, ventral view. Scale bars: 0.2 mm.

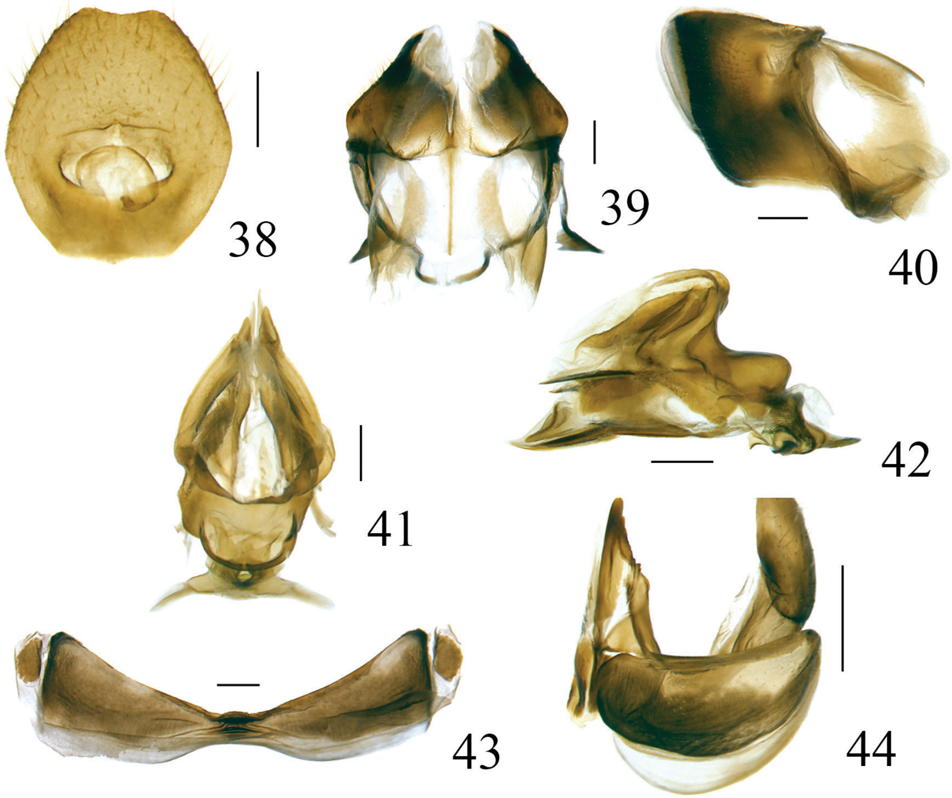


Figures 36–37. *Pusulissus coronomensis* sp. nov., male, holotype. **36** phallic complex, right lateral view **37** apex of phallic complex, posterior view. Abbreviations: dll: dorso-lateral lobe of periandrium; Ade: Aedeagus; vl: ventral lobe of periandrium; Adep: Aedeagus processes. Scale bars: 0.2 mm.

of female anal tube concave at middle (Fig. 38), while in *P. phiaoacensis* it is nearly straight (Fig. 15).

Description. Length: male (including forewings) (*N* = 2): 5.1–5.3 mm; female (including forewings) (*N* = 1): 5.3 mm.

Coloration. General appearance brown (Figs 25, 26). Vertex tawny, with two brown circular markings near the base, midline brown, margins black (Fig. 27). Compound



Figures 38–44. *Pusulissus coronomensis* sp. nov., female, paratype. **38** anal tube, dorsal view **39** gonoplacs, ventral view **40** gonoplacs, lateral view **41** gonapophysis IX and gonospiculum bridge, dorsal view **42** gonapophysis IX and gonospiculum bridge, lateral view **43** sternite VII, ventral view **44** gonocoxa VIII and gonapophysis VIII, lateral view. Scale bars: 0.2 mm.

eyes brownish dark, supported by tawny callus (Fig. 27). Frons brown, dorsal part black (Fig. 28); dorsal and lateral margins carinated by black, median carina brown (Fig. 28). Frons with two yellowish round markings in middle area, apical and lateral areas distributed with around twelve yellowish tubercles on each side, basal part yellow (Fig. 28). Antennae brown (Fig. 28). Postclypeus brown mixed with some tawny (Fig. 28). Genae tawny (Fig. 26). Pronotum tawny, median area with two brownish small impressions (Fig. 27), paranotal lobes brown mix with some yellow (Fig. 28). Mesonotum tawny (Fig. 27). Forewings tawny with veins tawny, the basal part of costal area with some irregular black markings, the middle area from basal third of costal margin extending to the middle of forewing surface also have some irregular black markings, the apical margin with a black linear marking (Fig. 29); some specimens with the black markings on forewing almost invisible, but with an obscure large paler round marking at basal third on each side of forewing (Fig. 25). Hindwings brown (Fig. 30). Legs tawny (Fig. 26).

Head and thorax. Vertex 2.3 times wider than long in midline, anterior margin straight, posterior margin angularly concave at middle, but the level shallower than the new species described above (Fig. 27). Frons 1.2 times longer in midline than the widest part, 1.2 times wider at widest part than dorsal margin (Fig. 28). Pronotum 1.1 times longer in midline than vertex in midline, lateral margins straight and oblique (Fig. 27). Mesonotum 2.2 times wider along anterior margin than long in midline (Fig. 27). Forewings twice longer at longest part than widest part (Fig. 29). Hindwing with apical margin of CuP-Pcu-A1 lobe rounded (Fig. 30). Metatibiotarsal formula: 2-(7–8)/(6–8)/2.

Male genitalia. Anal tube in dorsal view mushroom shaped, widest beyond middle, as long in midline as widest part, apical margin rounded (Fig. 32), lateral margins roundly convex in dorsal view (Fig. 32) and visible in lateral view (Fig. 31); anal opening located near the middle of anal tube (Fig. 32). Pygofer in lateral view subrectangular, dorsal margin slightly sloping to the posterior, dorso-lateral angle rounded, posterior margin almost parallel with the anterior margin (Fig. 31). Gonostylus in lateral view with dorsal margin elevated at the basal third (Fig. 33). Capitulum of gonostylus derived after the middle of gonostylus, broad, sharp triangular, directed to anterior; the meniscate processes with one side beyond the base of anterior margin of capitulum and another side beyond the posterior margin of capitulum (Fig. 33). In lateral view, periandrium dorso-lateral lobe and ventral lobe rounded apically (Figs 34, 36). In posterior view, ventral lobe of periandrium spinous protruded at middle in apical margin (Fig. 37). The lateral processes of aedeagus relatively short, hook-shaped, derived from the middle, directed anteriorly to basal fourth, reaching the ventral margin of periandrium (Figs 34, 36); in ventral view this pair of processes directed downwards (Fig. 35).

Female genitalia. Anal tube in dorsal view ovate, 1.1 times longer in midline than widest part, widest before mid length, apical margin slightly concave at middle, lateral margins rounded, anal opening situated slightly below middle (Fig. 38). Anterior connective lamina of gonapophysis VIII with two obscure large teeth in the apex but the keeled teeth in outer-lateral margin invisible (Fig. 44). Gonocoxa VIII long, quadrangular (Fig. 44). Hind margin of sternite VII roundly concave, the median part with a small convex protuberance (Fig. 43).

Etymology. Arbitrary euphonic name referring to the crown (latin 'corona') of yellow pustules on the frons.

Note. Genes sequences were registered in GenBank with the following accession numbers: MT772139 (whole 18S), MT772137 (28S D3–D5), MT772136 (28S D6–D7), MT774094 (COXI) and MT774093 (Cytb). For COXI of 681 bp length, this species differs by 57 bp with *P. phiaoacensis* and by 50 bp with *Pusulissus* sp. (Fig. 46).

Pusulissus gen. nov. distribution

With three different species, the new genus *Pusulissus* appears distributed around the South China in the Guangxi Province (Hezhou, Qichong Natural Reserve) and in North Vietnam (Cao Bang Province: Phia Oac and Vinh Phuc Province: Tam Dao)

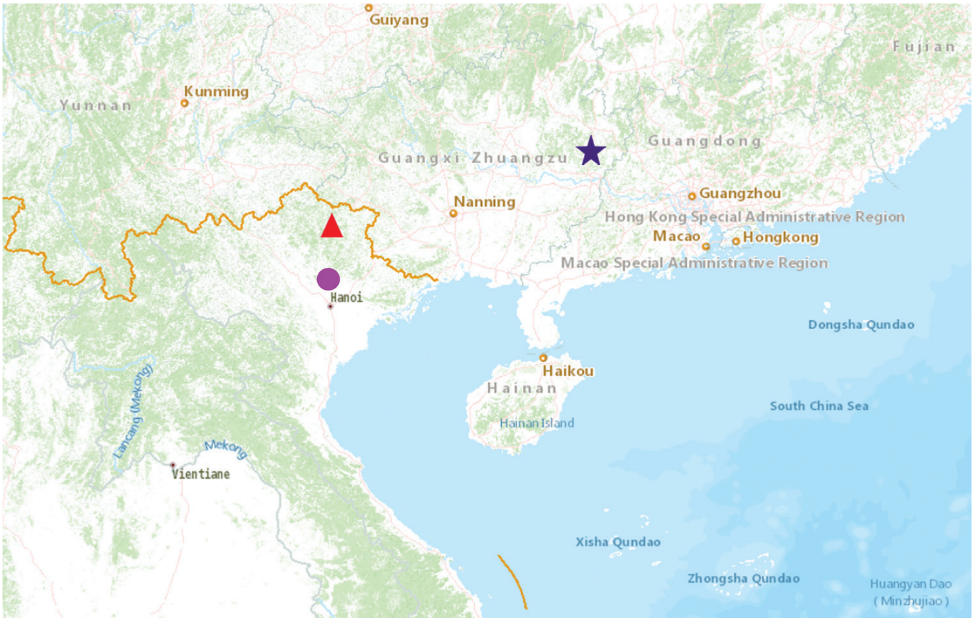


Figure 45. Distribution map of *Pusulissus*. The red triangular, blue five-pointed star and purple circular respectively indicate the distribution of species *P. phiaoacensis* sp. nov., *P. coronomensis* sp. nov. and *Pusulissus* sp.

(Fig. 45). The Vietnamese specimens were collected in mountainous biotopes at relatively high altitudes (between 750 and 1050 m) while at lower altitude (180 m) in China.

Pusulissus gen. nov. phylogeny

The new genus *Pusulissus* refers to the taxon “Gen. nov. *apud Tetricodes*” in Wang et al. (2016)’s molecular phylogenetic analyses, and three different species are observed in this study (Fig. 46). However, only two species are formally described here as the third taxon, *Pusulissus* sp., corresponding to the one already sequenced as “Gen. nov. *apud Tetricodes*” in Wang et al. (2016), is represented by only one incomplete female specimen from Vietnam also: Tam Dao, Vinh Phuc Province.

Within Parahiraciini, and from the combined genes (18S rRNA, 28S rRNA, COXI and Cytb) phylogeny (Fig. 46), the new genus *Pusulissus* appears sister to the genus *Tetricodes* Fennah, 1956, and both are sister to the genus *Tetricodissus* Wang, Bourgoin & Zhang, 2015 and *Neodurium* Fennah, 1956 in this study (Fig. 46), while this topology was ((*Pusulissus* + *Tetricodes*) + (*Tetricodissus* + *Neodurium*)) in Wang et al. (2016). *Pusulissus phiaoacensis* sp. nov., *Pusulissus* sp. and *Pusulissus coronomensis* sp. nov. respectively differ by 98 bp, 89 bp and 101 bp with the species *Tetricodes songae* Zhang & Chen, 2009 on a total of 681 bp in COXI.

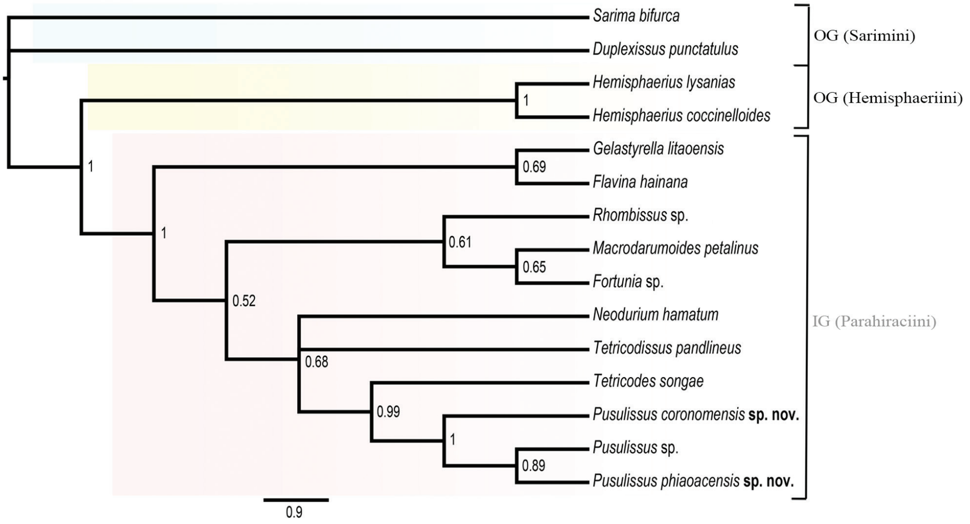


Figure 46. Bayesian 50% consensus tree of Parahiraciini species based on combined sequences (18S, 28S, COXI, Cytb) with 2 species of Sarimini and 2 species of Hemisphaeriini as outgroup. At each node, values denote posterior probability support.

Regard to the tribe Parahiraciini

The Parahiraciini lineage was erected within the Issidae by Cheng and Yang (1991a) for the single genus *Parahiracia* Ôuchi, 1940 with a subfamily rank. The subfamily recognition was based on the elongate ovate body, the absence of wax plates of abdominal segment VII-VIII (present in other Issidae sec. Fennah 1954) and the presence of respectively 10 and 8 median sensory pits on each side of the meso- and metanotum in the 5th instar larva, versus 2–5 and 0–2 sensory pits in other issids (Cheng and Yang 1991a, b). This lineage was confirmed as a monophyletic group by several successive molecular analyses (Wang et al. 2016; Bourgoin et al. 2018, unpublished data) while it was not recovered by Gnezdilov et al. (2020), probably due to methodological biases which is out of the scope of this paper.

From a morphological perspective and for the adults, Gnezdilov and Wilson (2007) characterized the tribe by 1) beetle-like conformation, convex, elongate, and apically narrowed forewings not exceeding the length of the abdomen with a net of apical transverse veins and 2) long fore and middle legs. Gnezdilov and Wilson (2007) added a “well-developed three- or two lobed (anal lobe more or less reduced) hindwings with a deep notch between remigium and vannus and a net of transversal veins in the distal part”, but regarded this last character as plesiomorphic. In their review and identification key to genera, Zhang and Chen (2012, 2013) used characters of the head capsule (presence or absence of swollen frons, vertex conformation), prothoracic femora and tibiae (flattened or not), forewing conformation and claval suture (present or not) and some genitalia characters. However, none of these characters appear as specific of the tribe. More recently, Gnezdilov (2017) added the “cuspidal apex of clavus” (Gnezdilov 2015) in the diagnosis of the tribe and also retained (Gnezdilov 2018) the narrow anal lobe, the deep cubital cleft, and the cuspidal apex of the forewing clavus as apomorphies of the tribe.

In 2016, Wang et al. (2016) proposed a new classification of the family, confirming Cheng and Yang (1991a)'s separation of Parahiraciini with the recognition of several other new tribes. They put in light the importance of the hindwing conformation in adults that appeared being well characteristic for each lineage. Since, our further studies of many Oriental issid genera have confirmed this view and a clearer figure for Parahiraciini taxa (including several yet non-described new genera) has now emerged. We can now provide a non-ambiguous morphological definition of the tribe Parahiraciini allowing us to review its composition and to divide the tribe into three subtribes: Scantiniina subtribe nov., Vindilisina subtribe nov., and the nominal one Parahiraciina Cheng & Yang, 1991 subtribe nov., although we don't exclude that the first two might represent in the future distinct lineages of higher range value. The following subtribal diagnoses are therefore proposed, mainly based on the hindwing conformation (Figs 47–52), with a key to Parahiraciini subtribes.

Parahiraciini Cheng & Yang, 1991

Type genus: *Parahiracia* Ôuchi, 1940 [syn. of *Fortunia* Distant, 1909 (Gnezdilov et al. 2004)]

Diagnosis (modified from Gnezdilov 2017, 2018). Body more or less flattened dorso-ventrally, frons often projecting, usually with a proboscis, forewings usually with keel-shaped veins, caudo-dorsal angle of clavus of forewings usually in shape of distinct triangular lobe (cuspidal apex of clavus: Gnezdilov 2015). Hindwing 2 or 3-lobate, strongly notched at CuP with CuP-Pcu-A1 lobe generally slightly wider than Sc-R-MP-CuA lobe; the two lobes almost the same length. Pcu and $A1_1$ merging or not in basal half of forewing. A2 lobe wide or reduced. Net of transverse veins present in Sc-R-MP-CuA lobe, or absent.

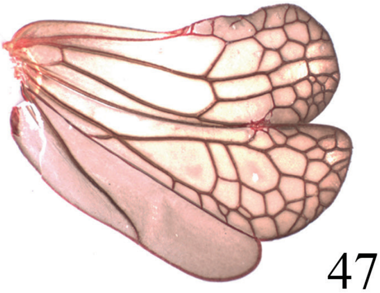
Parahiraciina Cheng &Yang, 1991 subtribe nov.

http://zoobank.org/84516219-0226-4258-BED7-A476E54DA37F
Figs 51, 52

Type genus. *Parahiracia* Ôuchi, 1940 [syn. of *Fortunia* Distant, 1909 (Gnezdilov et al. 2004)]

Diagnosis. The subtribe is identified and separated according to the following list of characters:

1. Hindwings bilobate, strongly notched at CuP with CuP-Pcu-A1 lobe generally wider than Sc-R-MP-CuA lobe; the two lobes almost the same length.
2. Posterior margin not or indistinctly notched at $A1_2$.



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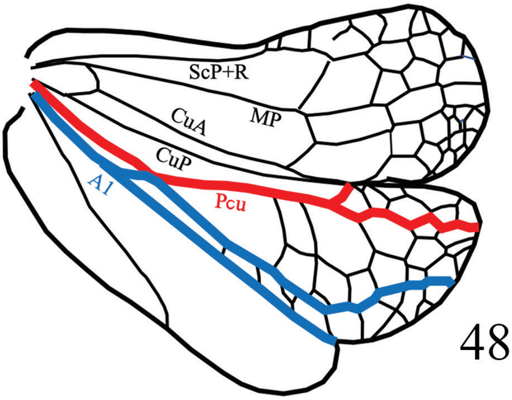

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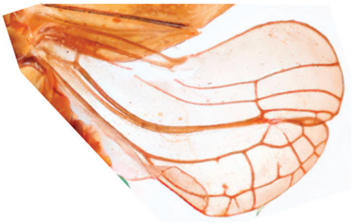

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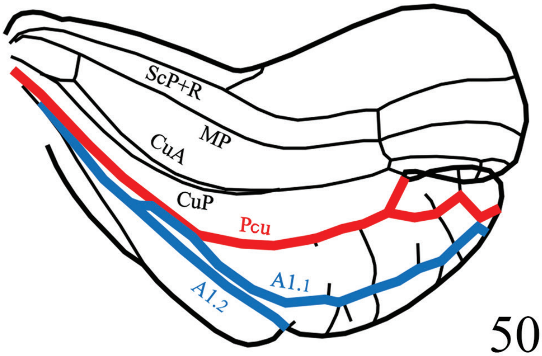

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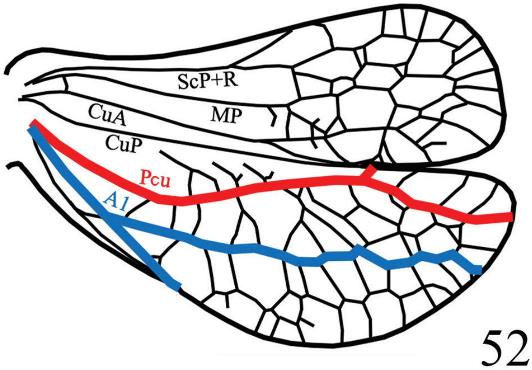

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Figures 47–52. Hindwing pattern and venation. **47, 48** Scantiniina subtrib. nov.: *Scantinius bruchoides* (Walker, 1858) (modified from Gnezdilov 2012, fig. 3) **49, 50** Vindilisina subtrib. nov.: *Vindilis fornicata* Stål, 1870 (modified from Gnezdilov 2018, fig. 3) **51, 52** Parahiraciina Cheng & Yang, 1991 subtrib. nov.: *Tetricodes tamdaoensis* Vanslembrouck & Constant, 2018 (modified from Vanslembrouck and Constant 2018, fig. 4).

3. A2 lobe with anal area posterior to A1 strongly reduced, much shorter and much thinner than the anterior lobes.
4. Sc-R-MP-CuA and CuP-Pcu-A1 lobes covered with a set of numerous transverse veins.
5. CuA and CuP not merging before the anterior notch.
6. Pcu and $A1_1$ not merging in basal half of forewing.
7. A2 present, not branched or absent. In some species a transverse a2-a1 connecting A2 with A1 at the level of its basal branching (*Tetricodes tamdaoensis* Vanslembrouck & Constant, 2018).

Note. Based on this diagnosis, Parahiraciina constitutes a well-defined group supported by several apomorphic characters (reduced anal lobe, numerous transverse veins) and the molecular analysis results. Accordingly, genera *Scantinius*, *Vindilis* and *Nisoprincessa* are moved to separate new subtribes. They probably all belong to the same higher lineage Parahiraciini based on the apomorphic strong cubital notch of the hindwing.

While *Folifemurum*, was already excluded from Parahiraciini by Wang et al. (2016), this view was not followed by Gnezdilov (2017). Based on its rounded hemisphaeriini general shape, its apomorphic one lobed hind wing, the medio-carinated frons, and particularly according to the molecular analysis result, it is here transferred to Hemisphaeriini Mongolianina. *Gelastyrella* is here maintained as a valid genus following Chen et al. (2014) versus Gnezdilov (2009)'s synonymy with *Thabena*, according to the number of small spines on the first metatarsi (more than 35 in *Gelastyrella* while less than 21 in *Thabena*), the large corpus connective of the phallic complex bearing a large and obvious ventrad expansion (corpus connective reduced and phallic complex without ventrad expansion in *Thabena*), and posterior margin of female sternite VII medially quadrate-shaped (triangular in *Thabena*) (Chen et al. 2014).

Scantiniina subtribe nov.

http://zoobank.org/D6FB9F06-AAE0-41F7-9DE2-9EB88A950398
Figs 47, 48

Type genus. *Scantinius* Stål, 1866.

Diagnosis. The subtribe is identified and separated according to the following list of characters:

1. Hindwing trilobate, strongly notched at CuA2 with CuP-Pcu-A1 lobe slightly longer, as wide as Sc-R-MP-CuA lobe.
2. Posterior margin distinctly notched at $A1_2$.
3. Anal lobe posterior to A1 present, surpassing half-length of CuP-Pcu-A1 lobe, apically rounded and 1/2 thinner than the anterior ones, and with margins sub-parallel.

4. Sc-R-MP-CuA and CuP-Pcu-A1 lobes covered with a set of numerous transverse veins.
5. CuA and CuP merging at the anterior notch.
6. Pcu and A1 merging in basal half of forewing.
7. A2 present, single.

Note. This subtribe is currently monogeneric and distributed in Indonesia (Sumatra), in Peninsular Malaysia and Sarawak (Gnezdilov and Wilson 2007). Based on the strong CuA2-CuP notch and the reticulated Sc-R-MP-CuA and CuP-Pcu-A1 lobes, it is tentatively considered as a sister lineage to Parahiraciina, but without molecular data, its place in the phylogeny of the Hemisphaeriinae sec. Wang et al. (2016) remains uncertain.

Vindilisina subtribe nov.

http://zoobank.org/FFC85FD7-1DD0-448D-9ABC-A064E19A4E30

Figs 49, 50

Type genus. *Vindilis* Stål, 1870.

Diagnosis. The subtribe is identified and separated according to the following list of characters:

Head capsule with vertex and frons in a slightly convex margin in lateral view. Compound eyes elongated, almost twice as long as wide in lateral view.

1. Hindwing trilobate, strongly notched at CuA2-CuP; Sc-R-MP-CuA and CuP-Pcu-A1 lobes almost the same wide.
2. Posterior margin distinctly notched at $A1_2$.
3. A2 lobe surpassing half-length of CuP-Pcu-A1 lobe, apically rounded and about 1/4 thinner than the anterior one, and with margins sub-parallel.
4. Sc-R-MP-CuA lobe not covered with a set of numerous transverse veins, a few incomplete ones in CuP-Pcu-A1 lobe.
5. CuA and CuP merging well before the anterior notch.
6. Pcu and A1 merging in basal half of forewing on some distance.
7. A2 present, single.

Note. The genera *Vindilis* and *Nisoprincessa* are transferred in this new subtribe, which is currently distributed only in Philippines (Palawan) (Gnezdilov 2017). The strong CuA-CuP notch looks similar to the other Parahiraciini, but both genera lack the reticulate venation of the Parahiraciina subtribe nov. or Scantiniina subtribe nov. In return, with exception of Hemisphaeriini, they share with Scantiniina subtribe nov. and other Hemisphaeriinae taxa the distinct plesiomorphic trilobate conformation of the hindwing. With Sarimini Wang, Zhang & Bourgoin, 2016 and Kodaianellini Wang, Zhang & Bourgoin, 2016, they exhibit the basal apomorphic merging of veins Pcu and A1. Without molecular data, its position in the phylogeny of Hemisphaeriinae remains uncertain and thus is currently left as a subtribe in the Parahiraciini.

Key to Parahiraciini subtribes

1 Hindwing trilobate, with a distinct notch at $A1_2$ vein apex separating a developed anal area apically widely rounded. Pcu and $A1_1$ merging in basal half of forewing **2**

– Hindwings bilobate, indistinctly notched at $A1_2$ apex; anal area short, triangular. Pcu and $A1_1$ not merging in basal half of forewing. Sc-R-MP-CuA and CuP-Pcu-A1 lobes covered with a set of numerous transverse veins (Figs 51, 52).. **Parahiraciina Cheng & Yang, 1991 subtribe nov.**

2 CuA and CuP merging at the anterior cubital notch. Sc-R-MP-CuA and CuP-Pcu-A1 lobes covered with set of transverse veins (Figs 47, 48)....... **Scantiniina subtribe nov.**

– CuA and CuP merging well before at the anterior cubital notch. Transverse veins almost absent in Sc-R-MP-CuA lobe (m-cu and r-m present), a few ones often incomplete in CuP-Pcu-A1 lobe (Figs 49, 50). Compound eyes elongated, almost twice as long as wide in lateral view............................... **Vindilisina subtribe nov.**

Conclusions

Parahiraciini constitutes a well-defined lineage in Issidae Hemisphaeriinae, easily recognized by the apomorphic cubital strong notch of the hindwing. Its monophyly is also fully supported by the molecular analyses at least for the nine genera, including *Pusulissus* gen. nov., described in this paper (Wang et al. 2016, Bourgoin et al. 2018, this paper). Paraphyly reported by Gnezdilov et al. (2020) in the maximum likelihood tree (fig. 2 pink box) is probably biased, as much as its Bayesian analysis also recovered a 100% supported node for the tribe (fig. 1). The three new subtribes proposed here are easily separated on morphological characters based on the hindwing conformation but their exact placement in the classification remains to be confirmed by molecular analysis. Within Parahiraciini, the placement of *Pusulissus* gen. nov. in the phylogeny in respective to the other Parahiraciina genera, remains of course provisional until a wider sampling would be available, but probably is close to the genus *Tetricodes*.

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Resin bees of genus *Megachile*, subgenera *Callomegachile* and *Carinula* (Hymenoptera, Megachilidae) from Thailand with description of a new species

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Abstract

Resin bees of the genus *Megachile* subgenus *Callomegachile* sensu lato (Hymenoptera; Megachilidae) from Thailand are reviewed. The 14 species treated include those described or revised in the subgenus *Alocanthedon*, a junior synonym of *Callomegachile* (three species), and in *Carinula* (one species). One new species is described, *Megachile chiangmaiensis* Chatthanabun and Warrit, **sp. nov.** The replacement name *Megachile parornata* Chatthanabun, Warrit and Ascher, **nom. nov.**, is proposed for *M. gigas* Wu (not Schrottky), which is recorded for the first time outside China. For each species, maps and full label data for the examined material documenting occurrences in Thailand are provided. In addition, global ranges, floral associations, and other life history data are summarized and a key to the Thai species is provided for females.

Keywords

Apoidea, Megachilini, Pollinator, Southeast Asia

Introduction

Bees in the subgenus *Callomegachile* Michener, 1962, of the genus *Megachile* Latreille, 1802 (both sensu lato of Michener 2007) are resin and mud-collecting megachilines native to the Old World that vary from moderate in size to gigantic for a bee (9.0–40.0 mm) and have an elongate, parallel-sided "chalicodomiform" body shape. *Callomegachile* are widespread in the Old World where native to the Paleotropics including all of sub-Saharan Africa, most of Asia extending northwards to subtropical and temperate latitudes, and the Australian region. At least 115 described species of *Callomegachile* sensu lato are currently known (Ascher and Pickering 2020; including 8 species of subgenus *Carinula* Michener, McGinley & Danforth, 1994) making it the most species-rich subgenera of resin bees and the second most species-rich overall (after the leafcutter bees subgenus *Eutricharaea* Thomson, 1872) in the genus *Megachile* (the third most species rich bee genus with 1490 valid species globally; Ascher and Pickering 2020), Most *Callomegachile* species collect plant resins as material for nest construction, hence the common name resin bees. In common with other species of Michener's (2007) *Megachile* Group 2, species of *Callomegachile* do not fold regularly-cut leaves to make their nests (a synapomorphy of his Group 1, corresponding to genus *Megachile* sensu stricto (Litman et al. 2011; Trunz et al. 2016; Gonzalez et al. 2019)), but some can incorporate irregularly-cut leaf pieces into nests, especially for making closures (Michener 2007).

In the New World, *Megachile* (*Callomegachile*) *umbripennis* Smith, 1853 is adventive on several Pacific Islands including the Hawaiian Islands and also locally in the Western Hemisphere (Michener 2007; Gonzalez and Engel 2012) where recently detected in South Florida (Ascher et al. 2016). *Megachile* (*Callomegachile*) *sculpturalis* Smith, 1853 of East Asia is now adventive in Eastern North America and widely distributed, having been found in southern Canada and in all of the Eastern United States (and in the Central States as far west as Nebraska, Kansas, and Texas). In addition, *Megachile* (*Callomegachile*) *rufipennis* (Fabricius, 1793) and *M.* (*Carinula*) *torrida* Smith, 1853 are adventive in the West Indies.

As with the delimitation of *Megachile* sensu lato, there has been considerable variation in subgeneric classification of *Callomegachile*, with Michener (2000, 2007) proposing a very inclusive concept of the group while noting the distinctiveness of several named lineages. However, other authors have partitioned these bees more narrowly, with Engel and Gonzalez (2011) describing a new subgenus *Alocanthedon* Engel & Gonzalez, 2011 based in part on clypeal shape in females and the presence of dense patch of black setae on male forewing. However, a recent molecular phylogenetic analysis (Trunz et al. 2016) treated *Alocanthedon* as a junior synonym of *Callomegachile*, which we follow here, pending more thorough investigation of relationships between the African and Asian taxa and, in particular, of the status of the gigantic *Megachile* such as *M. pluto* Smith, 1860 for which the name *Eumegachilana* Michener, 1965 is available (see Michener 2007).

In an analysis that resolved the traditional subgenus *Callomegachile* sensu lato (sensu Michener 2007) as polyphyletic, Trunz et al. (2016) found that *Carinula*, including the well-known Asian species *M. stulta* Bingham, 1897, were well separated from typical *Callomegachile*. For sake of completeness, we treat in this work all Thai resin bees including both *M.* (*Carinula*) and *M.* (*Callomegachile*) in the narrower sense, but with the former recognized as a separate subgenus in light of its divergent placement in Trunz et al.'s (2016) phylogenetic trees. It has distinctive characters such as a complete longitudinal median clypeal carina in females and lack of a front coxal spine in males (Michener 2007).

Morphological characters uniting *Callomegachile* sensu Michener (2007) include striate arrangement of punctures on mesoscutum and lower part of mesepisternum; mandibles of female bees usually equipped with 3 to 7 teeth with ridges that are minutely roughened (less so in *Carinula*, as noted by Michener 2007); small appressed hairs present on inner mandibular surface of adductor interspace. In males, the sixth metasomal tergum (T6) is weakly bilobed or lacks median emargination, and the gonoforcep is slender (broadened in *Carinula*) (Michener 1962, 1965, 2007).

Although many *Callomegachile* species are relatively well known and easy to recognize as such, more taxonomic and phylogenetic work is needed to clarify both subgeneric and species limits and to document newly discovered species. Many species described historically have poor original descriptions, are highly variable morphologically (especially in hair color), and have been inaccurately or controversially classified. In addition, the *Callomegachile* sensu lato fauna of Southeast Asia, including Thailand, remains poorly documented, in spite of their abundance and general distribution across the region (Gonzalez and Engel 2012; Ascher et al. 2016). Furthermore, interpretation of their biogeography has been complicated by their propensity to become adventive both across oceans and, potentially, within Asia itself (see Ascher et al. 2016; Soh et al. 2016).

Here, we summarize the occurrence of *Callomegachile* sensu lato species in Thailand from (1) literature records (2) historical specimens in two of the largest insect collection facilities in Thailand (3) the National University of Singapore, Division of Biological Sciences, Insect Diversity Lab database and (4) specimens recently collected from numerous collecting trips from 2006–2019. Distributions records, maps of Thai distributions, floral records, measurements, and images of pinned vouchers are provided based on study of both historical and recently collected specimens. Image records available for six species on the citizen science portal iNaturalist were also reviewed. One *Callomegachile* species new to science is described along with two new records for Thailand, and a replacement name is proposed for a species described from China and newly detected in Thailand.

Materials and methods

Within Thailand, 304 *Callomegachile* specimens (169♀, 135♂) were examined, which were deposited at the Natural History Museum of Chulalongkorn University (**NHM-CU-BSRU**; 147♀, 130♂), Bangkok, the Department of Entomology and Plant Pathology, Faculty of Agriculture, Chiang Mai University (**CMU**; 5♀), Chiang Mai, the Department of National Parks, Wildlife and Plant Conservation (**DNP**; 4♀), Bangkok, the Kasetsart Kamphaeng Saen Insect Collection (**KKIC**; 13♀, 5♂), Nakhon Pathom, Thailand. Additional specimens were examined in the Insect Diversity Lab Collection of the National University of Singapore including material on loan from the Oberösterreichisches Landesmuseum of Linz, Austria (curator Fritz Gusenleitner, including material assembled by Maximilian Schwarz). Type repositories used for comparison with specimens examined in this study are abbreviated as follows:

IZB	Chinese Academy of Science, Yunnan, China
MFNB	Museum für Naturkunde, Berlin, Gemany
NHMUK	The Natural History Museum, London, United Kingdom
NMNH	National Museum of Natural History, Smithsonian's Institution Washington DC, USA
USA–SEMC	Snow Entomological Museum Collection, Lawrence
ZMUC	Zoological Museum of the University of Copenhagen, Copenhagen, Denmark

Types of the valid species-group taxa were examined to the extent possible, including those of some but not at all of the names in synonymy. Image records on the citizen science portal iNaturalist were reviewed and identified by JSA.

Specimens were examined and measured with a Zeiss Stemi 508 dissecting microscope equipped with an ocular micrometer or calipers. Body length was measured from edge of clypeus (in dorsal view) to apex of T6. Forewing length was measured from tegula to lateral wing margin. Interocellar distance (ID) and ocelloccipital distance (OD) were measured and these distances were calculated into ID/OD proportion. Male genitalia were dissected following a method modified from Gonzalez (2008). Photomicrographs were prepared using a Canon 7D Mark II digital camera attached to a T2-T2 1.6× SLR long-distance microscope lens, and were processed with Adobe Photoshop CS6. Measurements are reported in millimeters using Axiovision LE 4.8.2.0. Bees were placed in an insect relaxing jar for 3–4 days to soften the specimens for facilitating the examination of mandibles and labrums. Mandibular teeth are numbered sequentially starting from apex toward the base of the mandible. Morphological terminology follows that of Michener and Fraser (1978), Engel (2001), and Michener (2007).

Specimens from NHMCU-BSRU were collected during 1956–1971 (33♀, 4♂) mostly by Dr. Kloom Vajropala of NHMCU-BSRU and 2006–2019 (112♀, 126♂) from recent surveys by the Thai authors. Label data including localities were recorded verbatim for each specimen and, following georeferencing if necessary, were used to construct distribution maps using Adobe Illustrator. Coordinates for the maps were generated based on specimen labels or, for records lacking GPS data, by georeferencing localities using Google Earth and GPS Geoplaner online. To obtain data on global distribution by country and primary subdivisions, literature records were critically reviewed from Tadauchi and Tasen (2009), Ascher et al. (2016), and the Discover Life Bee Species Guide and World Checklist (Ascher and Pickering 2020), and new records obtained were validated for future inclusion in the latter source. Type localities and repositories are cited, with details provided for those species described from Thailand including new records.

Taxonomy

Genus *Megachile* Latreille

Megachile Latreille, 1802: 413, 433. Type species: *Apis centuncularis* Linnaeus, 1758, by designation of Curtis, 1828, pl. 218.

Subgenus *Callomegachile* Michener

Chalicodoma (*Callomegachile*) Michener, 1962: 21. Type species: *Chalicodoma mystaceana* Michener, 1962, by original designation.

Chalicodoma (*Eumegachilana*) Michener, 1965: 191. Type species: *Megachile clotho* Smith, 1861, by original designation.

Chalicodoma (*Morphella*) Pasteels, 1965: 537. Type species: *Megachile biseta* Vachal, 1903, by original designation.

Chalicodoma (*Orientocressoniella*) Gupta, 1993: 165. Type species: *Megachile relata* Smith, 1879, by original designation [but Gupta's description refers to a bee quite different from the nominal type species, see Baker and Engel 2006].

Chalicodoma (*Alocanthedon*) Engel & Gonzalez, 2011: 53. Type species: *Chalicodoma odontophorum* Engel, 2011, by original designation.

Diagnosis. Body elongate. Female mandible three to seven teeth with minutely roughened ridge. Punctures on mesoscutum and lower part of mesepisternum striated. Small appressed hairs on inner mandibular surface of adductor interspace present. In males, carina on T6 bilobed or lacks median emargination; posterior margin of T6 simple and without tooth.

Comments. The subgenus *Pseudomegachile* Friese, 1898 is superficially similar to *Callomegachile*, in term of size and appearance. Females of *Pseudomegachile* can be recognized by the presence of long erected hairs on inner mandibular surface of adductor interspace, whereas males can be easily recognized by the presence of multidentate apical margin of T6.

Megachile (*Callomegachile*) *atratiformis* Meade-Waldo, 1914

Figs 1, 2

Megachile atratiformis Meade-Waldo, 1914: 456; Female syntype (NHMUK, examined) Mergui (Low Tenasserim), Myanmar.

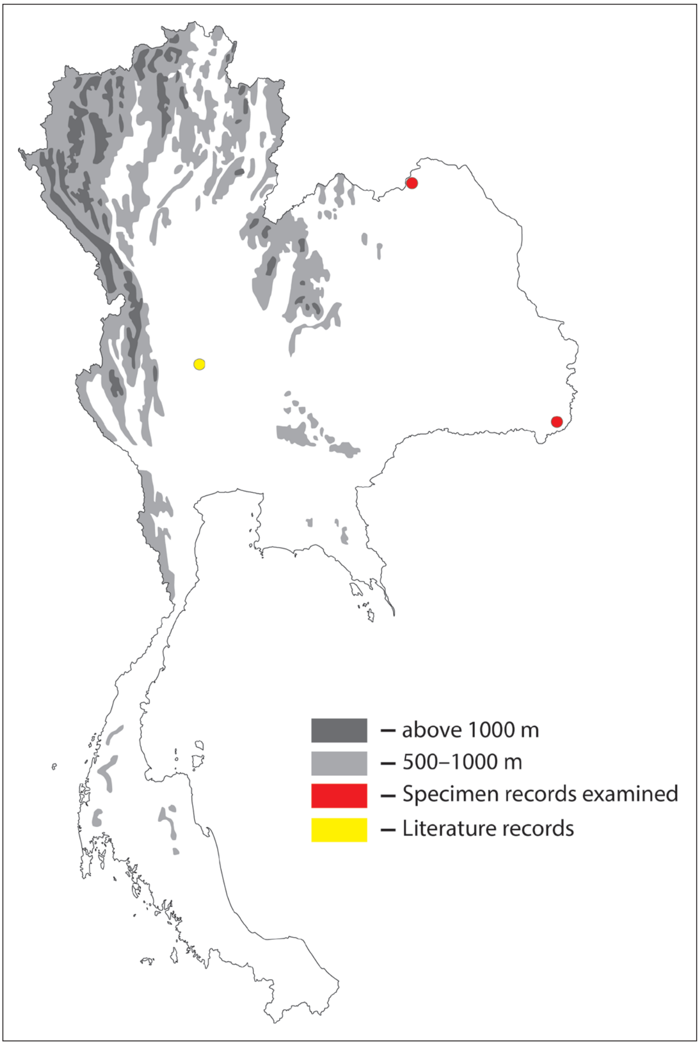


Figure 1. Thai distribution of *Megachile* (*Callomegachile*) *atratiformis*.

Chalicodoma (*Alocanthedon*) *atratiforme* Engel & Gonzalez, 2011: 68–70.

Diagnosis. Female can be easily recognized by its large body size (18–22 mm); black body covered with black hairs throughout (Fig. 2a); clypeus without median tubercle; mandibles four teeth; apical margin of labrum truncate without tooth (Fig. 2d); mesoscutum with weak transverse wrinkle pattern on disc, posteriorly also with weakly transverse wrinkle pattern (Fig. 2e); yellow wings; black scopa. Male can be easily recognized by the presence of black setae patch on medial cell of forewings; apical of T6 with shallow concavity; front tibia modified (Engel and Gonzalez 2011).

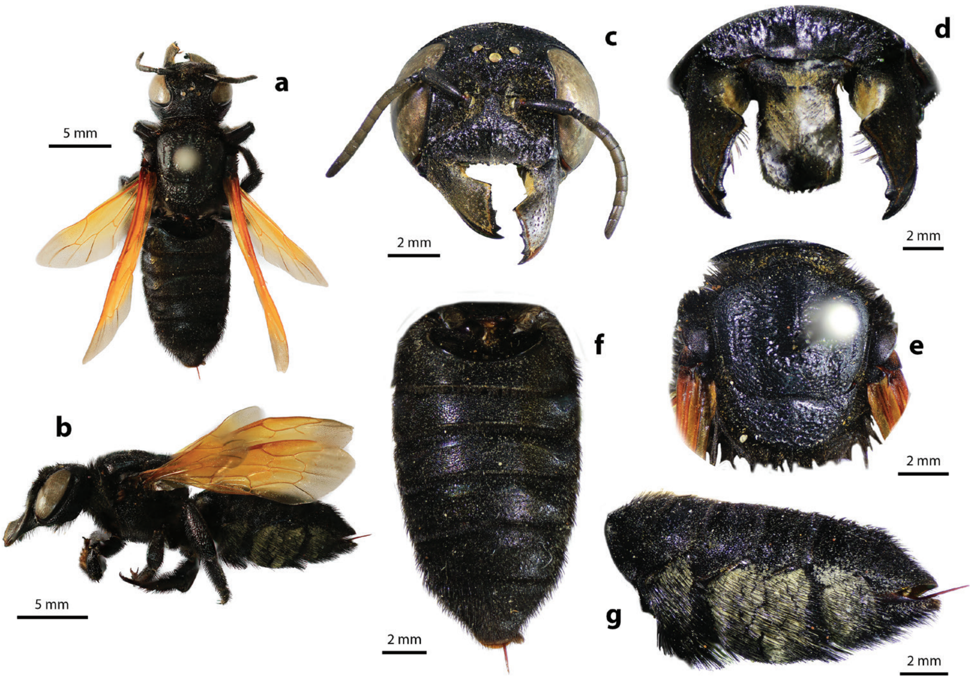


Figure 2. *Megachile* (*Callomegachile*) *atratiformis* Meade-Waldo, 1914, female **a** dorsal view **b** lateral view **c** frontal view **d** frontal view of mandible and labrum **e** dorsal view of mesoscutum **f** dorsal view of metasoma **g** lateral view of metasoma and scopa.

Literature records. Malaysia. Negeri Sembilan, Pahang (Meade-Waldo 1914; Gonzalez and Engel 2011; Ascher and Pickering 2020); Myanmar. Mergui Archipelago in Tanintharyi Region (Meade-Waldo 1914; Gonzalez and Engel 2011; Ascher and Pickering 2020); Thailand. Uthai Thani (Engel and Gonzalez 2011).

Material examined. Female syntype. Myanmar: Tannintharyi Region. "Type H. T.; B.M. TYPE HYM. 2037. 17. a.; *Megachile atrata* (Var), Smith, female; Lower Tenasserim. Mergui, 11–89. C. T. Bingham.; Col. C. T. Bingham 96–30; *Megachile* (*Eumegachile*) *atratiformis* M.W., G. Meade-Waldo det., Type, female; NHMUK 013379843"; Thailand. Nong Khai Province: 1♀, Phon Phisai district, Cowboy Coffee, 18°6'9.95"N, 103°6'17.96"E, Alt. 147.65 m, 15-I-2017, coll. N. Warrit et al. (leg. NC and NW). Ubon Ratchathani Province: 1♀, Phu Jong Na Yoi Nat. P., Kaeng Ka Lao, 14°26'10.98"N, 105°16'37.05"E, Alt. 322 m, 5-I-2019, coll. N. Warrit et al (leg. NC and NW).

Floral records. Engel and Gonzalez (2011) noted the species was captured on an indigenous tree, *Dipterocarpus obtusifolius* Teijsman & Miquel.

Comments. Meade-Waldo's original material was composite (Engel and Gonzalez 2011) so we exclude from the distribution records from Middle Tenasserim: Haundraw [also known as Houngdarau] Valley in Myanmar (= *M. odontophorum*) and also from Penang in Malaysia.

***Megachile* (*Callomegachile*) *disjuncta* (Fabricius, 1781)**

Figs 3–5

Apis disjuncta Fabricius, 1781: 481. Female type (ZMUC, not examined) "America" [erroneous; surely from Asia, and perhaps from India].
Anthophora disjuncta: Fabricius, 1804: 374.
Trachusa disjuncta: Jurine, 1807: 251.
Megachile disjuncta: Lepeletier, 1841: 331.
Megachile (*Pseudomegachile*) *disjuncta*: Friese, 1911: 207.
Chalicodoma (*Callomegachile*) *disjuncta*: Michener, 1965: 191.

Diagnosis. Female can be recognized by its medium body size (13–18 mm); black body covered with black hairs throughout, except propodeal triangle and T1 with white hairs (Fig. 4a); apical margin of clypeus with two small tubercles (Fig. 4d); mandibles five teeth with two stout apical teeth at apex and three small teeth basally; labrum rectangle; apical margin of clypeus truncate with two lateral teeth (Fig. 4e); wings hyaline except fuscous apical part; black scopa (orange in part in females of the superficially similar leafcutter bee *M.* (*Aethomegachile*) *conjuncta* Smith, 1853 which also has a shorter and less parallel-sided metasoma, finer tergal punctures, and cutting edges on mandibles). Male is similar to female except paraocular area and apical margin of clypeus with white hairs (Fig. 5c); mandibles three teeth; labrum rectangle with round corners (Fig. 5d).

Literature records. China. Anhui, Beijing, Fujian, Guangxi Zhuang, Guizhou, Hainan, Hebei, Hunan, Jiangsu, Jiangxi, Shandong, Shanghai, Shanxi, Sichuan, Zhejiang; India. Andaman and Nicobar Islands: Long Island (notable records from Cockerell 1912), Andhra Pradesh, Chandigarh, Haryana, Karnataka, Maharashtra, Puducherry, Tamil Nadu, Uttarakhand (Lepeletier 1841; Smith 1873; Gribodo 1884; Bingham 1897; Cameron 1907; Friese 1911; Cockerell 1912; Cockerell 1919; Flectcher 1920; Dover 1921; Schulthess-Rechberg 1935; Ascher et al. 2016); Indonesia. Java, Sumatra (Gribodo 1884; Schulthess-Rechberg 1935; Ascher et al. 2016); Madagascar. Antananarivo, Bourbon, Toamasina (Gribodo 1884; Grandidier 1890; Pauly 2001; Ascher et al. 2016); Malaysia. Kelantan, Kepong, Kuala Lumpur, Negeri Sembilan, Penang, Selangor, (Ascher et al. 2016); Mauritius. Rodrigues Island (Smith 1873; Gribodo 1884; Bingham 1897; Cameron 1907; Pauly 2001; Ascher et al. 2016); Myanmar. Tenasserim (Bingham 1897; Flectcher 1920; Dover 1921; Schulthess-Rechberg 1935); Reunion. (Pauly 2001; Ascher et al. 2016); Seychelles. Mahe, Morne Blanc, Praslin Island (Cameron 1907; Cockerell 1912; Pauly 2001; Ascher et al. 2016); Singapore. (Ascher et al. 2016); Sri Lanka. Hambantota, Puttalam (Ascher et al. 2016); Vietnam. Thai Binh (Ascher and Pickering 2020). Additional records based on images on iNaturalist are from Bangkok, Chaiyaphum, Surat Thani, and Udon Thani Provinces (credit: lennyworthington 2017; nopcoeur 2018; bernhard_hiller 2019; tonykris 2019; alexeyreshchikov 2020).

Material examined. Thailand. Ayutthaya Province: 1♀, XI-1961, coll. unknown (leg. NC and NW); 1♀, Beung Pra ram, 01-VIII-1970, coll. unknown (leg. NC and

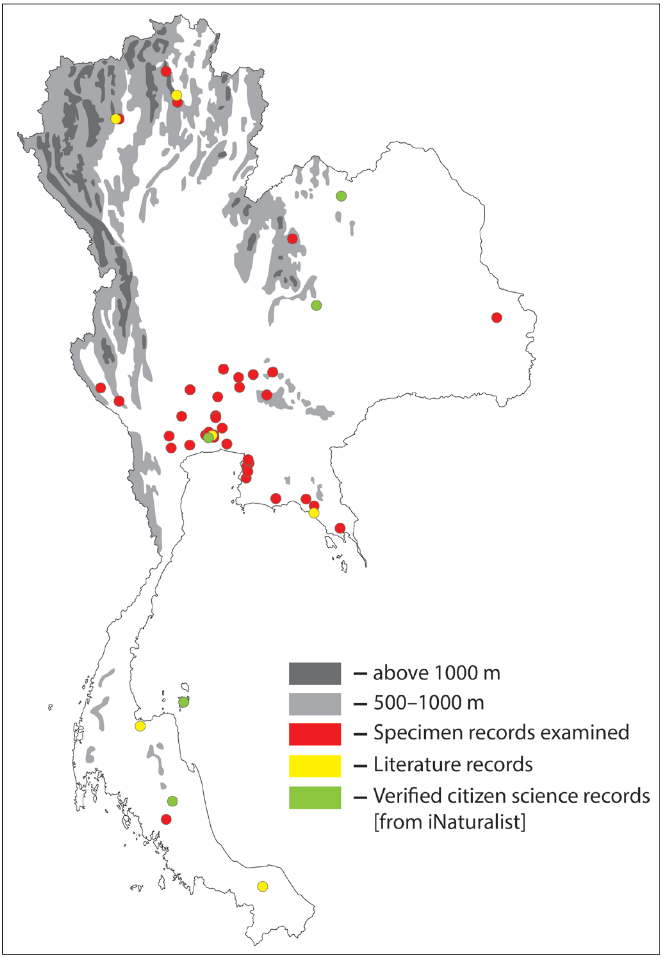


Figure 3. Thai distribution of *Megachile* (*Callomegachile*) *disjuncta*.

NW). Bangkok Province: 1♀, Phra Nakhon, 05-I-1960, coll. unknown (leg. NC and NW); 1♀, Phra Nakhon, 12-VII-1960, coll. unknown (leg. NC and NW); 1♀, 15-II-1963, coll. unknown (leg. NC and NW); 1♀, Nong Khaem district, 16-IX-1966, coll. Wanida (leg. NC and NW); 1♀, Bang Khen district, 19-VII-1969, coll. unknown (leg. NC and NW); 1♀, Thonburi district, 01-VII-1970, coll. unknown (leg. NC and NW); 1♀, Sa Torn district, Thungmahamek subdistrict, 22-IV-1971, coll. unknown (leg. NC and NW); 1♀, 05-VIII-1971, coll. unknown (leg. NC and NW); 1♀, Sathorn district, Soi Suanglu1, 05-X-2009, coll. K. Attasopa (leg. NC and NW); 1♀, Sathorn district, Soi Suanglu1, 06-X-2009, coll. K. Attasopa (leg. NC and NW). Chanthaburi Province: 1♀, Tha Mai district, 19-IX-1969, coll. unknown (leg. NC and NW); 1♀, Makam district, 25-III-2015, coll. N. Chatthanabun (leg. NC); 1♀, 08-V-1956, coll. unknown (leg. NC and NW); 1♀, Bang Pra district, 08-

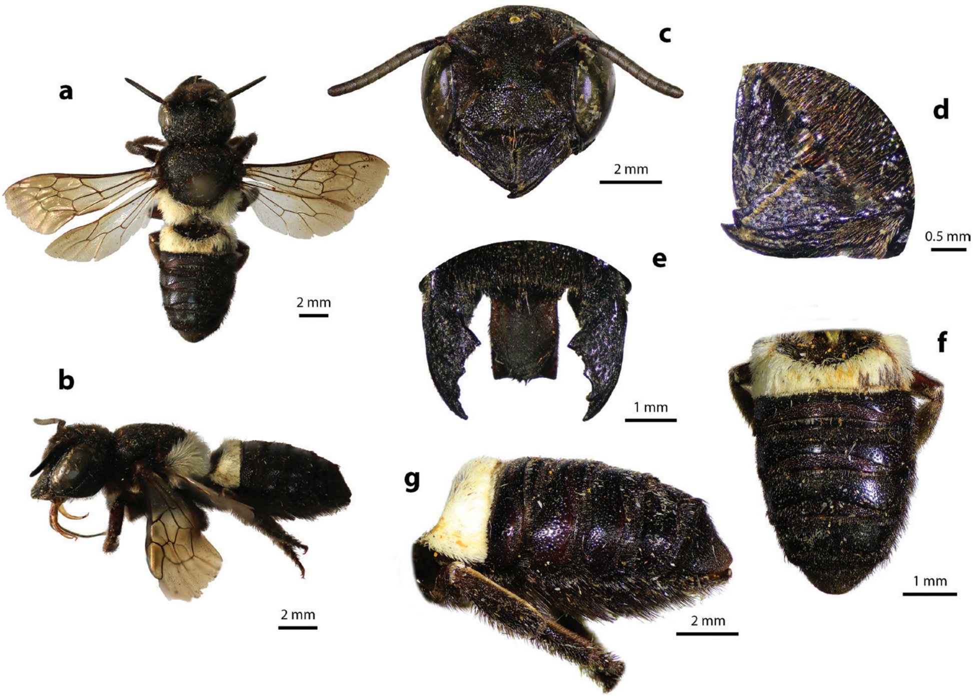


Figure 4. *Megachile* (*Callomegachile*) *disjuncta* (Fabricius, 1781), female **a** dorsal view **b** lateral view **c** frontal view **d** oblique view of clypeal margin **e** frontal view of mandible and labrum **f** dorsal view of metasoma **g** lateral view of metasoma and scopa.

X-1963, coll. unknown (leg. NC and NW); 1♀, Ang Sila, 04-VII-1970, coll. Nonglak (leg. NC and NW); 1♀, Sriracha district, Bang Pra Reservoir, 04-VII-1971, coll. unknown (leg. NC and NW); 1♀, Mueang district, Soi Na Khao Bor Yang, 13°6'10.2066"N, 100°57'59.5764"E, Alt. 15.86 m, 27-VII-2017, coll. P. Traiyasut (leg. NC and NW). Chiang Mai Province: 1♀, 09-VII-1959, coll. Unknown (leg. NC and NW); 39 ♀, Mueang district, Suthep subdistrict, Faculty of Agriculture Chiang Mai University, 18°47'38.6772"N, 98°57'32.9220"E, Alt. 391 m, 19-VII-2015, coll. Warrit et al. (leg. NC and NW); 17♂, Mueang district, Mae Hia subdistrict, 18°45'51.1272"N, 98°55'39.6192"E, Alt. 232 m, 19-VII-2015, coll. Warrit et al. (leg. NC and NW). Chiang Rai Province: 1♀, Pan district, 17-I-2009, coll. T. Yusing (leg. NC and NW). Loei Province: 1♀, Phu Kradueng district, Phu Kradueng National Park, 16°52'22.4934"N, 101°50'11.7384"E, Alt. 506.28 m, 29-V-2016, coll. Warrit et al. (leg. NC and NW). Kanchanaburi Province: 12♂, Sai Yok district, Wang Krachae subdistrict, 14°9'56.7678"N, 99°3'30.5640"E, Alt. 101.80 m, 24-VI-2016, coll. Warrit et al. (leg. NC and NW); 2♂, Sai Yok district, Wang Krachae subdistrict, 14°11'6.5724"N, 99°3'6.9258"E, Alt. 102.30 m, 24-VI-2016, coll. Warrit et al. (leg. NC and NW). Lop Buri Province: 1♀, 10-IX-2013, coll. Warrit (leg. NC and NW). Nakhon Nayok Province: 1♀, Khao Yai National Park, 06-IX-1970, coll. unknown

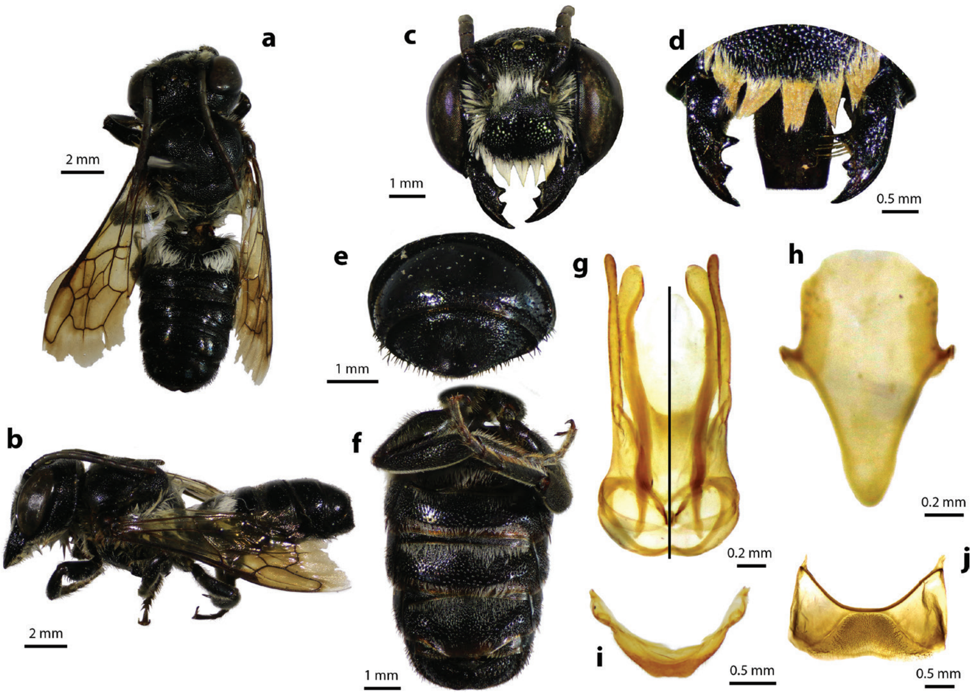


Figure 5. *Megachile* (*Callomegachile*) *disjuncta* (Fabricius, 1781), male **a** dorsal view **b** lateral view **c** frontal view **d** frontal view of mandible and labrum **e** frontal view of T7 **f** ventral view of metasomal sterna **g** dorsal (left) and ventral (right) views of penis **h** dorsal view of S8 **i** T7 **j** S5.

(leg. NC and NW). Nakhon Pathom Province: 1♂, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 06-II-2008, coll. Patneti (leg. NC and NW); 1♂, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 15-VI-2011, coll. Patneti (leg. NC and NW); 1♀, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 30-XI-2013, coll. Sunita (leg. NC and NW); 1♀, 4♂, Kamphaeng Saen district, 13°44'58.3908"N, 99°52'33.1242"E, Alt. 14 m, 10-VII-2015, coll. Warrit et al. (leg. NC and NW); 1♀, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 26-I-2016, coll. Noppasiri (leg. NC and NW); 1♀, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 20-III-2016, coll. K. Laesen (leg. NC and NW); 1♀, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 29-III-2016, coll. Jirawat (leg. NC and NW); 1♀, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 14-IV-2016, coll. Adisak (leg. NC and NW); 1♂, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 11-V-2016, coll. Vilunda (leg. NC and NW); 1♂, Kamphaeng Saen district, Kasetsart University Kamphaeng Saen campus, 04-IX-2016, coll. S. Laengsaruk (leg. NC and NW); 5♀, Kamphaeng Saen district, 14°0'35.1072"N, 100°0'35.9028"E, Alt. 11 m, 12-II-2017, coll. N. Chatthanabun, N. Warrit and V. Sivayyapram (leg. NC and NW). Nakhon Ratchasima Province:

1♀, Pak Chong, 06-I-1964, coll. unknown (leg. NC and NW); 1♀, Pak Chong, 28-VII-1969, coll. W. Sooksri (leg. NC and NW). Nonthaburi Province: 1♀, Pak Kred district, Khlong Phapha, coll. unknown (leg. NC and NW). Pathum Thani Province: 1♂, Ban Ngew district, Samkok subdistrict, Pai rom temple, 18-II-1968, coll. Chairoj (leg. NC and NW); 1♂, 18-II-1968, coll. Rojnee (leg. NC and NW). Phayao Province: 1♂, Mueang district, Mae Ka subdistrict, Phayao University, 03-I-2009, coll. Saowalak (leg. NC and NW); 16♂, Mueang district, Mae Ka subdistrict, Phayao University, 01-VI-2012, coll. Warrit et al. (leg. NW); 1♀, Mae Ka, Phayao University, 19°2'45.3510"N, 99°52'40.5438"E, Alt. 464.26 m, 5-VII-2016, coll. Warrit et al. (leg. NC and NW). Ratchaburi Province: 1♀, 22-VIII-1970, coll. unknown (leg. NC and NW); 1♀, 17-VIII-1973, coll. Pinpong (leg. NC and NW); 2♀, Potharam district, 13°44'50.0070"N, 99°53'38.4180"E, Alt. 6 m, 13-VII-2015, coll. Warrit et al. (leg. NC and NW); 1♀, 01-V-2017, coll. Pornchanok (leg. NC and NW). Rayong Province: 1♀, Ban Pae, 03-VIII-1971, coll. unknown (leg. NC and NW). Samut Prakan Province: 1♂, Park Nam district, 11-VIII-1963, coll. P. Sakulmon (leg. NC and NW). Samut Sakhon Province: 1♂, Banpaew district, 19-VIII-2014, coll. P. Tangtorwongsakul (leg. NC and NW). Saraburi Province: 1♀, 30-VIII-1960, coll. B. Pasook (leg. NC and NW); 1♀, Muak Lek, 03-VIII-1967, coll. unknown (leg. NC and NW); 1♀, Phu Khae, 04-II-1968, coll. Decha (leg. NC and NW); 1♀, Muak Lek, 03-VIII-1971, coll. Unknown (leg. NC and NW). Suphan Buri Province: 1♀, 13-IV-2015, coll. Kaewkan (leg. NC and NW). Trat Province: 1♀, Mueang district, Ao Yai, 23-III-2015, coll. N. Chatthanabun (leg. NC). Trang Province: 1♂, Na Yong district, 7°33'8.0892"N, 99°46'33.6072"E, Alt. 24 m, 11-VI-2015, coll. Warrit et al. (leg. NC and NW). Ubon Ratchathani Province: 1♂, Trakan Phuetphon district, Sarin lake view village, 03-VIII-2014, coll. N. Chatthanabun (leg. NC). Unknown localities: 1♂, 16-VIII-1960, coll. unknown (leg. NC and NW); 1♀, 01-I-1965, coll. unknown (leg. NC and NW); 2♀, 2♂, coll. unknown (leg. NC and NW).

Floral records. Throughout Thailand, *M.* (*Callomegachile*) *disjuncta* can be found abundantly in many agricultural plots that are planted with *Crotalaria juncea* L., a common plant species grown for providing essential nitrogen element to many crop plants in Thailand. Also, a common weed, *Bidens pilosa* L., is frequently visited by the species. One record of *M.* (*Callomegachile*) *disjuncta* was found on *Cratoxylum cochinchinense* (Lour.) Blume.

Comments. Female somewhat superficially resembles *M.* (*Aethomegachile*) *conjuncta* in size and overall appearance.

***Megachile* (*Callomegachile*) *faceta* Bingham, 1897**

Figs 6–8

Megachile faceta Bingham, 1897: 486. Female syntype (NHMUK, examined) Pegu Hills, Myanmar.

Megachile faceta rufojugata: Cockerell, 1931: 2.

Chalicodoma (*Callomegachile*) *faceta*: Michener, 1965: 191.

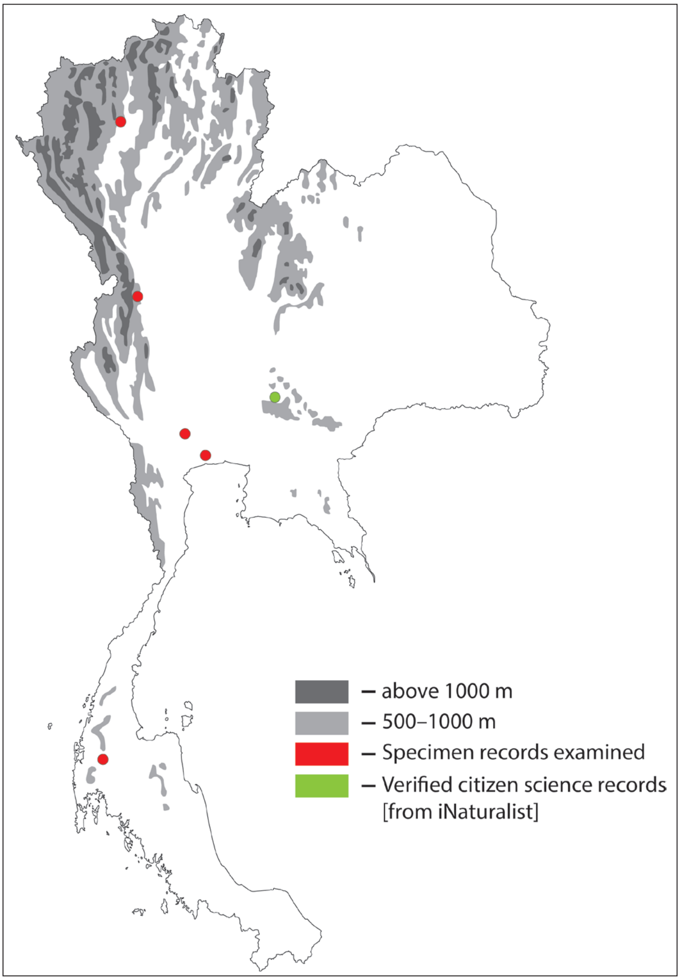


Figure 6. Thai distribution of *Megachile* (*Callomegachile*) *faceta*.

Diagnosis. Female can be recognized by its black medium body size (12–14 mm); vertex and pronotum covered with fulvous hairs; propodeal triangle and T1–T4 with tuft white hairs on lateral edges; apical margin of T5 with white hair band but interrupted at median (Fig. 7a); apical margin of clypeus with two small tubercles (Fig. 7c); mandibles five teeth with two stout apical teeth at apex and three small teeth basally; labrum rectangle with rounded corners (Fig. 7d); vertex with median carina (Fig. 7e); white scopa except black at apical area. Male is similar to female except mandible three teeth; labrum rectangular with shallow impression at median (Fig. 8d).

Literature records. India. Khasia Hills (Cockerell 1911); Myanmar. Pegu Hills [as Bago Yoma], Tanintharyi Region [as Tenasserim] (Bingham 1897); Taiwan. Sauter (Cockerell 1911). An iNaturalist image shows four females among a group of bees photographed at Hin Tung, Mueang District, Nakhon Nayok Province (credit: scottyastro 2015).

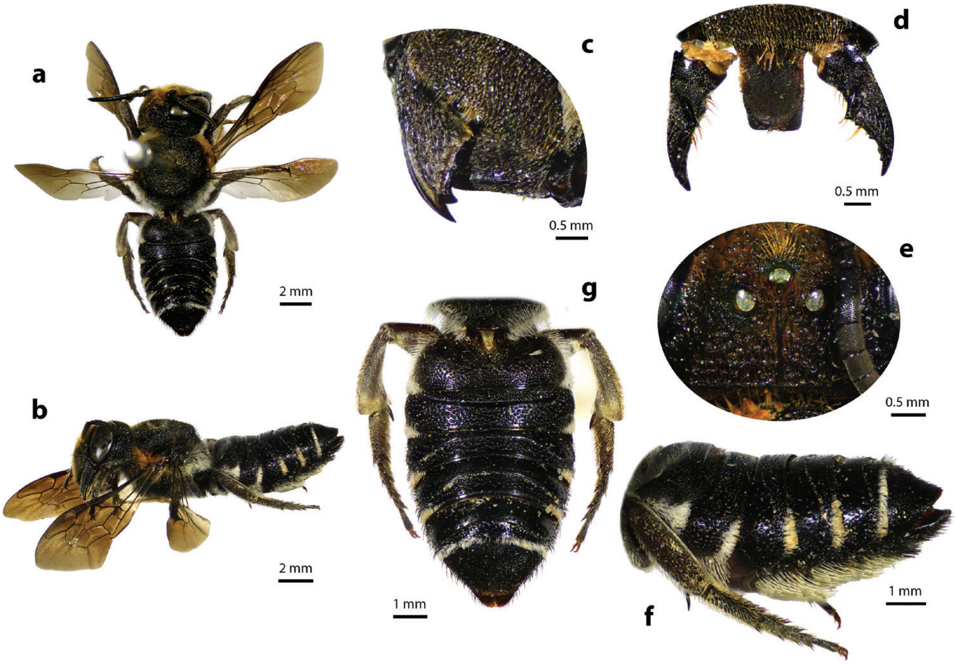


Figure 7. *Megachile* (*Callomegachile*) *faceta* Bingham, 1897, female **a** dorsal view **b** lateral view **c** oblique view of clypeal margin **d** frontal view of mandible and labrum **e** dorsal view of median carina at vertex **f** lateral view of metasoma and scopa **g** dorsal view of metasoma.

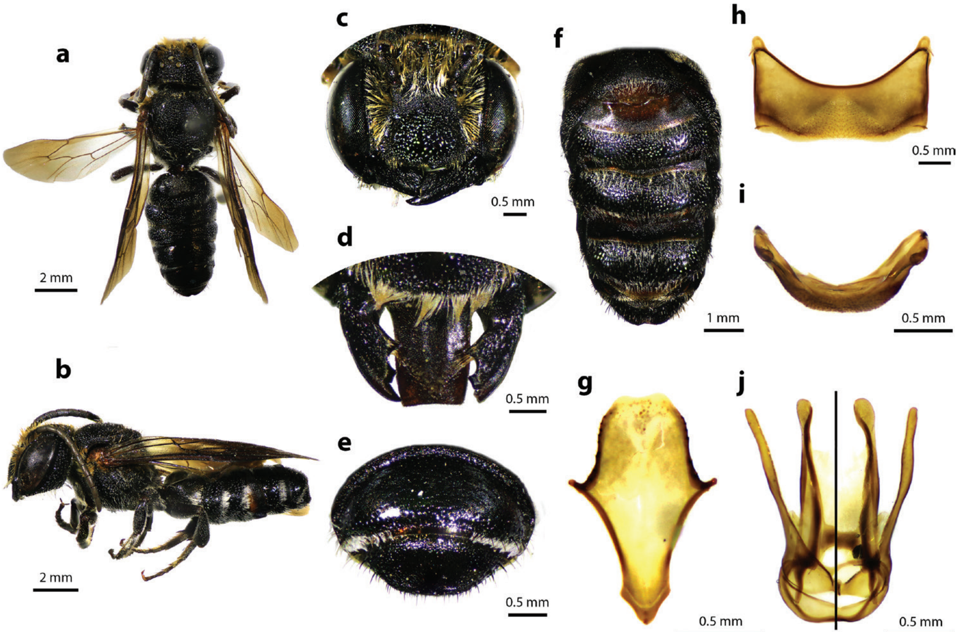


Figure 8. *Megachile* (*Callomegachile*) *faceta* Bingham, 1897, male **a** dorsal view **b** lateral view **c** frontal view **d** dorsal view of mandible and labrum **e** frontal view of T7 **f** ventral view of metasomal sterna **g** dorsal view of S8 **h** S5 **i** T7 **j** dorsal (left) and ventral (right) views of penis.

Material examined. Female syntype. Myanmar. "Type; B.M. TYPE HYM. 2011 17. a.; *Megachile faceta* Bingh. Female, Type; Pegu Hills, Burma, 11-87, Bingham Coll; Col. C. T. Bingham 96–30; NHMUK 013379842"; Thailand. Bangkok Province: 1♀, Pom Prap Sattru Phai district, Khlong Mahanak subdistrict, Sapan Kaw, 18-VII-1971, coll. Sudthida (leg. NC and NW). Chiang Mai Province: 13♂, Mueang district, Mae Hia subdistrict, Mae Hia Agricultural Research, Demonstrative and Training Center, 18°45'51.1272"N, 98°55'39.6192"E, Alt. 232 m, 19-VII-2015, coll. Warrit et al. (leg. NC and NW). Nakhon Pathom Province: 1♀, Kamphaeng Saen district, Kamphaeng Saen subdistrict, Kasetsart University, Kamphaeng Saen Campus, 02-IX-2010, coll. Pakkawat (leg. NC and NW). Nakhon Sawan Province: 2♀, Mae Wong Natural Park, 28-VI-2015, coll. V. Sivayyapram (leg. NC and NW). Surat Thani Province: 1♀, Phanom district, 8°54'35.9460"N, 98°31'37.9590"E, Alt. 118.68 m, 27-I-2018, coll. Warrit et al. (leg. NC and NW).

Comments. The similar *Megachile* (*Callomegachile*) *facetula* Cockerell, 1918, described from Sandakan, Sabah, Borneo, should be looked for in Thailand, but we have not been able to confirm any records. *Megachile strupigera* Cockerell, 1922, from Canton (now Guangzhou in Guandong) in southern China is likely a junior synonym of *M. faceta* based on our examination of images of its type in the NMNH, whereas Wu (2006) placed it in the leafcutter subgenus *Amegachile* Friese, 1909. Multiple species of leafcutter bees present in the region including Thailand closely resembles *M. facetula* in color pattern, so all identifications must be considered structural characters as well.

***Megachile* (*Callomegachile*) *fulvipennis* Smith, 1879**

Figs 9, 10

Megachile fulvipennis Smith, 1879: 68. Female holotype (NHMUK, examined) Nicobar Island, India.
Megachile atratiformis sininsulae Cockerell, 1927: 160.
Chalicodoma (*Callomegachile*) *atratiforme sininsulae*: Michener, 1965: 191.
Chalicodoma (*Callomegachile*) *fulvipennis*: Michener, 1965: 191.

Diagnosis. Female superficially resembles *M.* (*Callomegachile*) *atratiformis* (Meade-Waldo, 1914), *M.* (*Callomegachile*) *memecylonae* (Engel, 2011) and *M.* (*Callomegachile*) *odontophora* (Engel, 2011), in overall appearance: black body covered with black hairs throughout; yellow wings; black scopa (Fig. 10a, f) except smaller size (14–16 mm); punctures on mesoscutum and lower part of mesepisternum striated; mandibles five teeth with two stout apical teeth at apex and three small teeth basally; labrum rectangular (Fig. 10c).

Literature records. India. Andaman and Nicobar Island (Smith 1879; Ascher et al. 2016); Indonesia. Java, Sumatra (Ascher et al. 2016); Malaysia. Perak, Selangor, Terengganu (Ascher and Pickering 2020); Singapore. (Ascher et al. 2016).

Material examined. Female holotype. Nicobar Island [India]. "Holotype; B.M. TYPE HYM. 2055 17. a.; *Megachile fulvipennis* Sm. (Type); Nicobar, 76–30; NHMUK 013379844"; Thailand. Nakhon Pathom Province: 1♀, Kamphaeng Saen

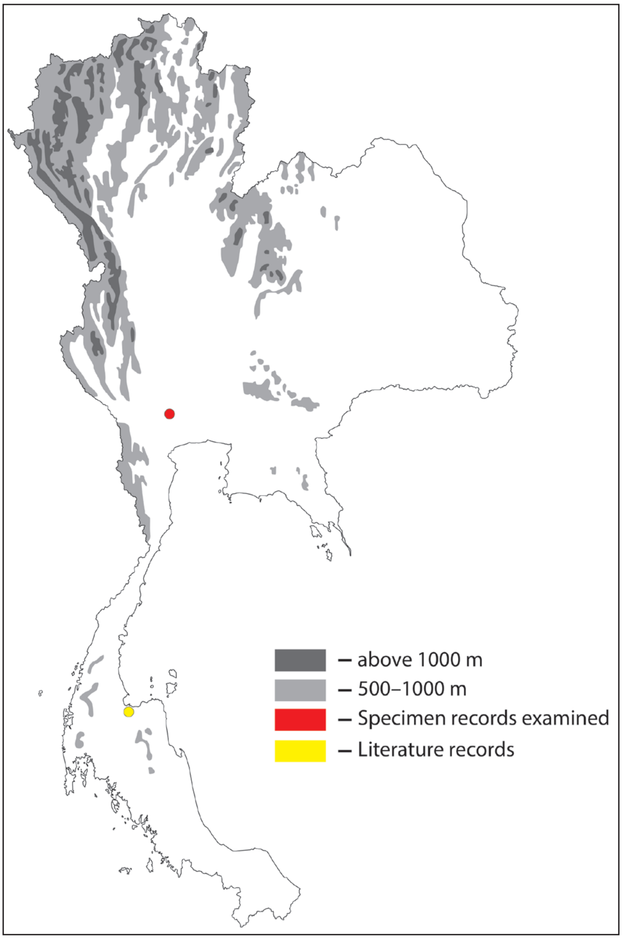


Figure 9. Thai distribution of *Megachile* (*Callomegachile*) *fulvipennis*.

district, Kamphaeng Saen subdistrict, Kasetsart University, Kamphaeng Saen Campus, 20-VIII-1956, coll. Chayuta (leg. NC and NW).

***Megachile* (*Callomegachile*) *impressa* Friese, 1903**

Figs 11, 12

Megachile impressa Friese, 1903: 358. Male holotype (MFNB, not examined) Tenasserim (Kayin: Thandaung), Myanmar.

Diagnosis. The species superficially resembles *M.* (*Callomegachile*) *binghami* Meade-Waldo, 1912, in terms of its overall appearance and size: black hairs on paraocular area;

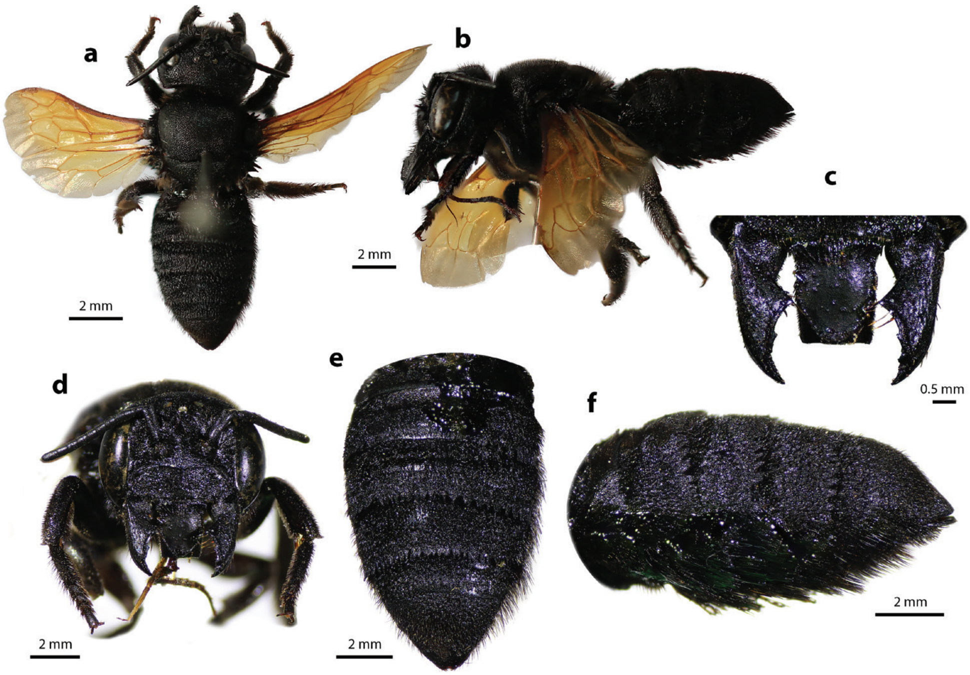


Figure 10. *Megachile* (*Callomegachile*) *fulvipennis* Smith, 1879, female **a** dorsal view **b** lateral view **c** dorsal view of mandible and labrum **d** frontal view **e** dorsal view of metasoma **f** lateral view of metasoma and scopa.

thorax with white hairs except central area of mesoscutum and scutellum; metasomal terga covered with ferruginous hairs; scopa ferruginous except white basal area; apical margin of clypeus with one medioapical tubercle and two lateral tubercles (Fig. 12b, c); meso- and metatarsi with ferruginous hairs (Fig. 12a). These characters are used to associate male and female bees.

Description. ***Female.*** Length. Total body length 14.28–15.90; wingspan 23.40–26.54; fore wing 10.01–11.34. Structure and color. Head black; paraocular area with dense black hairs; central area of clypeus with strong median carina; apical margin of clypeus with medioapical tubercle and two lateral tubercles; subtriangular supraclypeal area with sparse punctures, apical and median area with strong carina; mandible with two stout apical teeth at apex and three small teeth basally, without cutting edge; outer surface of mandible minutely roughened with long black hairs; labrum rectangular, with surface minutely roughened and brimmed with erected long brown hairs along margins, conspicuously at apex; gena with sparse punctures; almost bare vertex with sparse punctures, ID shorter than OD, ID/OD = 0.50 ± 0.01; antennae with ten flagella, first flagellomere wider than long but shorter than the second; body parallel-sided, thorax covers with white hairs except central area of mesoscutum and scutellum; mesoscutum and lower part of mesepisternum with coarsely striate puncture pattern; procoxal base with conspicuous small carina, covered with sparse white hairs; pro- and

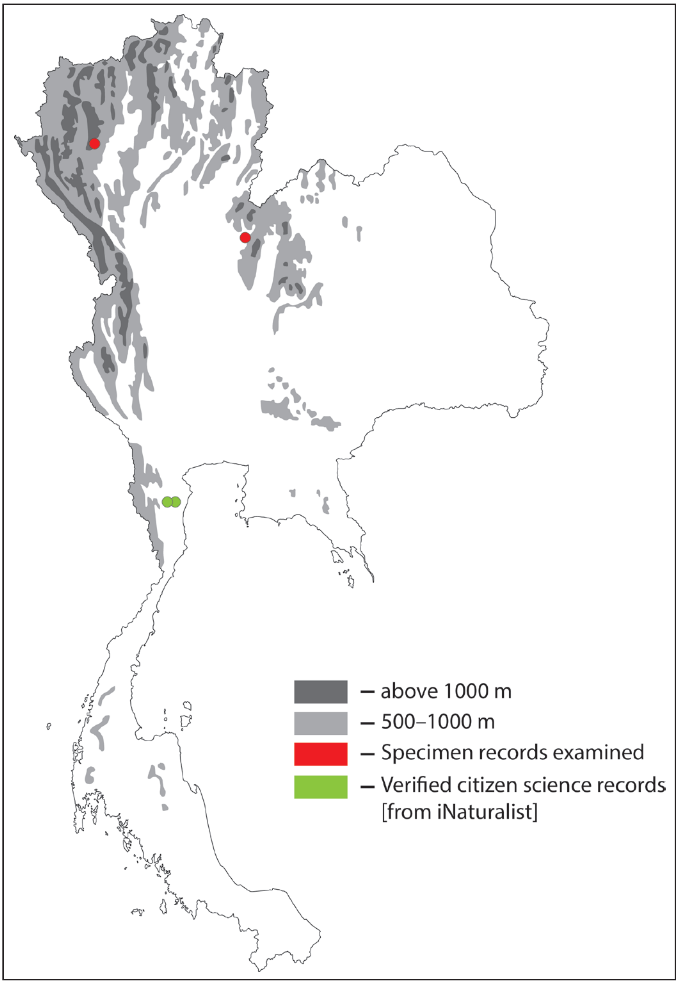


Figure 11. Thai distribution of *Megachile* (*Callomegachile*) *impressa*.

mesotibiae with two spines at apices; metatarsus with one small spine at apex; protarsus covers with dense brown hairs; meso- and metatarsus cover with dense brown hairs on outer side, with dense ferruginous hairs inner side; wing brown with dark brown vein; T1 covers with sparse ferruginous hairs, with sparse punctures; T2–T5 cover with dense ferruginous hairs, dense punctures on pregradular area, sparse punctures on marginal zone; T6 covers with dense ferruginous hairs, with sparse punctures and round shape at apex; scopa ferruginous except the basal area with white.

Literature records. Laos. Houaphan (Ascher and Pickering 2020); Malaysia. Kelantan (Ascher and Pickering 2020); Myanmar. Kayin, Tangdong, Tenasserim. (Friese 1903; Ascher and Pickering 2020). Also recorded based on images on iNaturalist on two occasions from Huai Mae Priang, Kaeng Krachan District, Phetchaburi Province (credit: djhiker 2016; pam-piombino 2017).

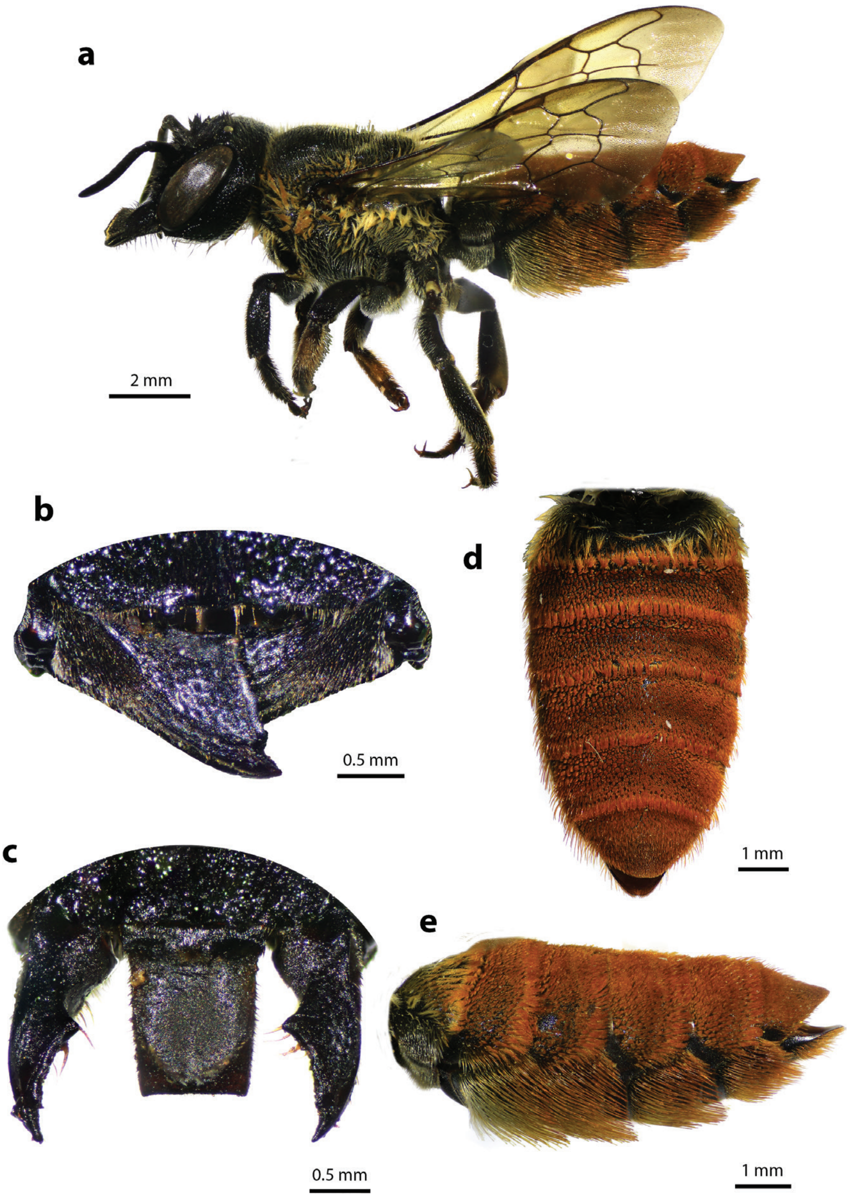


Figure 12. *Megachile* (*Callomegachile*) *impressa* Friese, 1903, female **a** lateral view **b** frontal view **c** clypeus and mandible **d** dorsal view of metasoma **e** lateral view of metasoma.

Material examined. Thailand. Chiang Mai Province: 1♀, Chom Thong district, Doi Inthanon Nat. P., Wildfire Control Station, 18°37'7.0590"N, 98°36'29.7606"E, Alt. 779 m, 20-VII-2015, coll. Warrit et al. (leg. NC and NW). Phitsanulok Province: 2♀, Phuhinrongkla Nat. P., 16°59'49.3008"N, 101°0'40.6772"E, Alt. 1303 m, 17-VI-2017, coll. N. Warrit et al. (leg. NC and NW).

Floral record. Specimens from Phitsanulok province were collected on *Craspedolobium unijugum* (Gagnepain) Z. Wei & Pedley at Phuhinrongkla National Park along with *M. umbripennis* and *Megachile* (*Creightonella*) *fraterna* Smith, 1853.

Comments. In the field, *M. impressa* can be confused with *Megachile* (*Creightonella*) *fraterna* since that species also has a black head and ferruginous hairs on abdomen. However, *M. fraterna* can be easily discriminated by characters in the mandible and cutting edge between interspaces.

Remarks. Known sites for this species are in the highlands.

***Megachile* (*Callomegachile*) *memecylonae* (Engel, 2011)**

Figs 13, 14

Chalicodoma (*Alocanthedon*) *memecylonae* Engel, 2011: 63–67; Male holotype (NHMUK, examined), Penang: Batu Ferringgi, Malaysia.

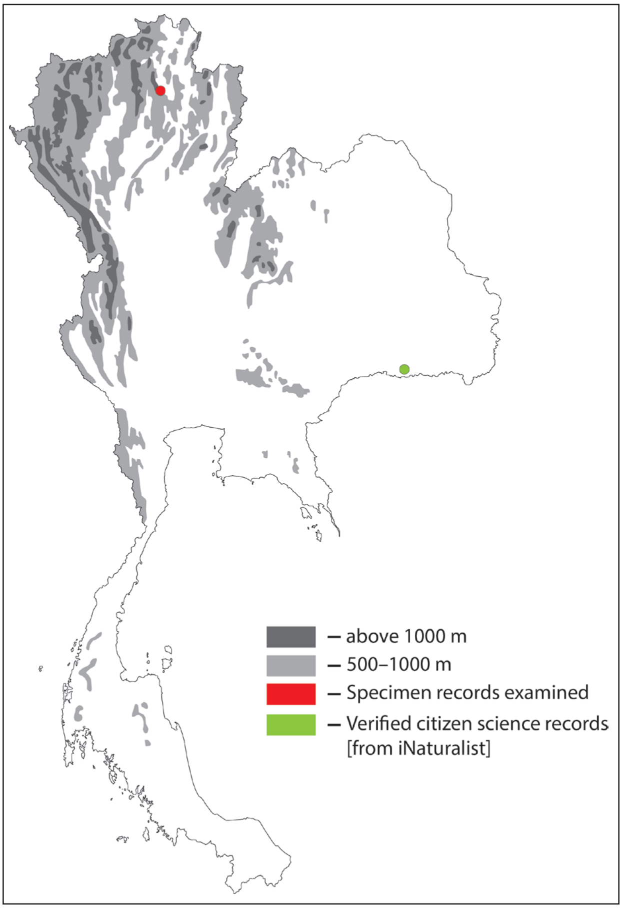


Figure 13. Thai distribution of *Megachile* (*Callomegachile*) *memecylonae*.

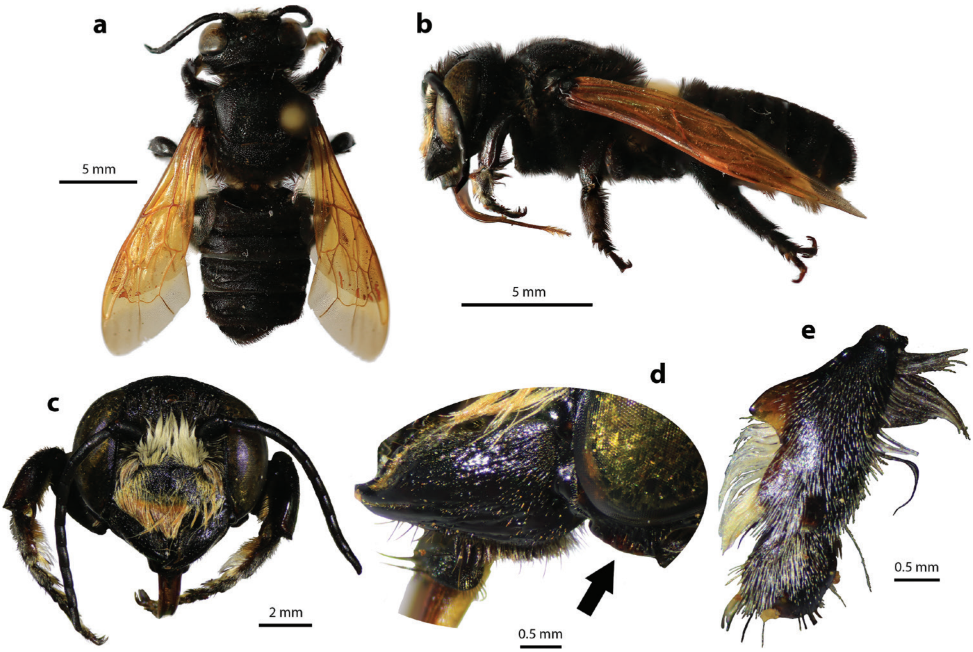


Figure 14. *Megachile* (*Callomegachile*) *memecylonae* (Engel, 2011), male **a** dorsal view **b** lateral view **c** frontal view **d** juxtamandibular flange **e** basitarsi of front leg.

Diagnosis. *Megachile* (*Callomegachile*) *memecylonae* (Engel, 2011) superficially resembles *M.* (*Callomegachile*) *atratiformis* (Meade-Waldo, 1914), in overall appearance and size: large body size (18–19 mm); black body covered with black hairs throughout; yellow wings. Female is easily distinguished by mesoscutum with distinctly transverse wrinkle pattern on disc, posteriorly also with well separated punctures (Engel and Gonzalez 2011). Male can be recognized by median cell of forewings without black setae patch; juxtamandibular flange present (Fig. 14d); basitarsi of pro legs with hook shape (Fig. 14e).

Literature records. Malaysia. Kuala Lumpur, Pahang, Pangkor Island, Penang, Perak, Selangor (Engel and Gonzalez 2011). A photograph on iNaturalist from Buachet, Surin Province may pertain to this species but visual identification in this group is difficult (credit: janescan 2018).

Material examined. Male holotype. Malaysia. "Holotype; B.M. TYPE HYM 17a. 3179; memecylonae, Batu Feringgi; 17 xi 1963 HTP. ø147; H.T. Pagden Coll. B.M. 1971–46; Malaya, Penang, Batu Feringgi, 17 Nov 1963, H T Pagden; Holotype, *Chalicodoma memecylonae* Michael S. Engel; NHMUK 013380270"; Thailand. Phayao Province: 1♂, Mueang district, Mae Ka subdistrict, Phayao University, 01-VI-2012, coll. Warrit et al. (leg. NW).

Remarks. This species was described from Peninsular Malaysia. It is remarkable that the first and only Thai specimen record is from Phayao Province in northern Thailand.

***Megachile* (*Callomegachile*) *monticola* Smith, 1853**

Figs 15, 16

Megachile monticola Smith, 1853: 179. Female syntype (NHMUK, examined).
Megachile felderi Radoszkowski, 1882: 79.
Megachile rhinoceros Mocsáry, 1892: 131.
Megachile samson Cameron, 1897: 128.
Megachile koshunensis Strand, 1913: 60.
Chalicodoma (*Eumegachilana*) *monticola*: Michener, 1965: 192.

Diagnosis. Female can be recognized by its black large body size (20–26 mm); mesosoma and T1 covered with fulvous hairs (Fig. 16a); base of clypeus with large protruding tubercle (Fig. 16b); mandibles elongate with three teeth and small tubercle at base; labrum oblong with lateral impression (Fig. 16c); black scopa.

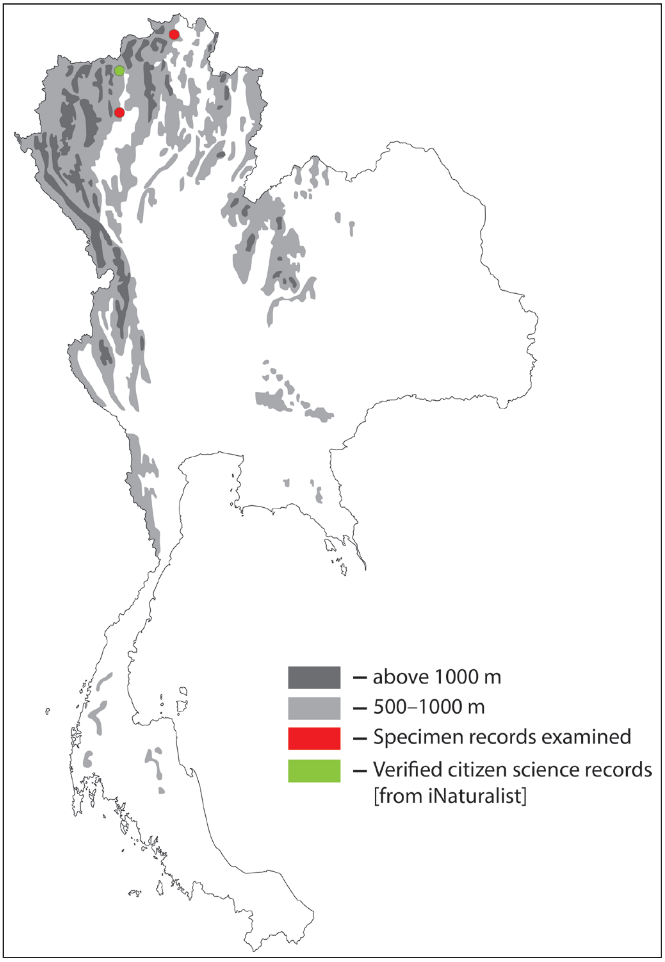


Figure 15. Thai distribution of *Megachile* (*Callomegachile*) *monticola*.

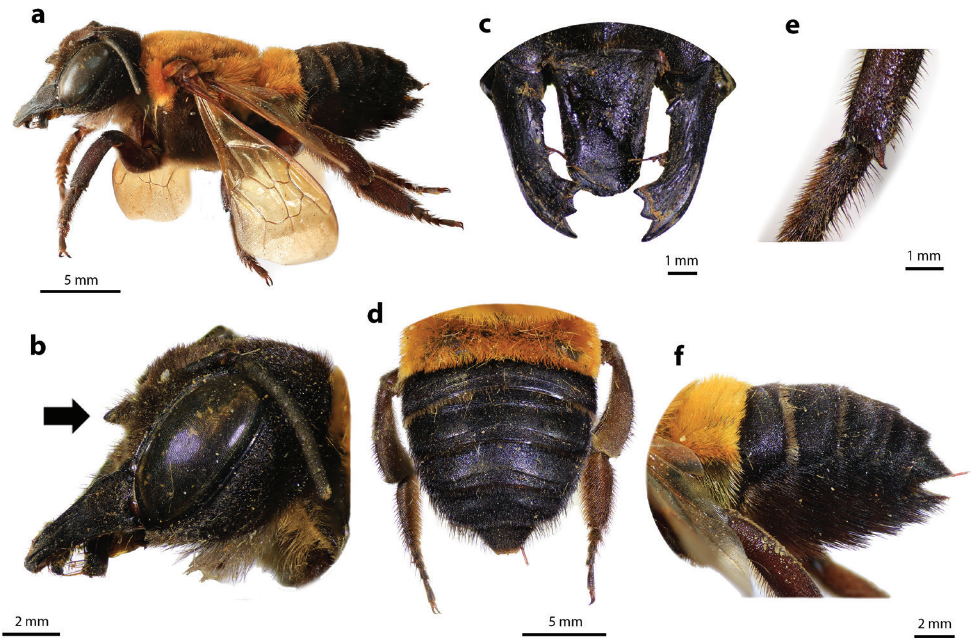


Figure 16. *Megachile* (*Callomegachile*) *monticola* Smith, 1853, female **a** lateral view **b** protruding tubercle at base of clypeus (arrow) **c** frontal view of mandible and labrum **d** dorsal view of metasoma **e** spine at apical tibia **f** lateral view of metasoma and scopa.

Literature records. Bangladesh. Sylhet (Smith 1853; Bingham 1897); China. Anhui, Fujian, Shanghai (Smith 1853); Hong Kong.; India. Assam, Sikkim (Bingham 1897); Indonesia. Lombien Island (Ascher and Pickering 2020); Japan. Okinawa, Uragami (Ascher and Pickering 2020); Myanmar. Tenasserim (Bingham 1897); Taiwan. Taihoku-shu (Ascher and Pickering 2020); Vietnam. Thua Thien-Hue (Ascher and Pickering 2020). Also reported from Chiang Mai Province on iNaturalist (credit: entomokot 2019).

Material examined. Female syntype. "Type; B.M. TYPE HYM. 17. a. 2155; Sylhet, 4[?]7 51; *Megachile monticola*, TYPE, Sm.; monticola Type Sm.; NHMUK 013379845"; Thailand. Chiang Mai Province: 1♀, Mueang district, Chang Phuag subdistrict, Nong Hoe, 17-VII-1996, coll. Adul (leg. NC and NW); 1♀, longan plantation, 10-II-2009, coll. Paveenun (leg. NC and NW); 1♀, 20-IX-1985, coll. Sumrid (leg. NC and NW). Chiang Rai Province: 1♀, Mae Chan district, Mae Chan subdistrict, 14-VIII-1960, coll. unknown (leg. NC and NW).

Megachile (*Callomegachile*) *odontophora* (Engel, 2011)

Figs 17, 18

Chalicodoma (*Alocanthedon*) *odontophorum* Engel, 2011: 55–60; Female paratype (NHMUK, examined) Nakhon Ratchasima, Thailand.

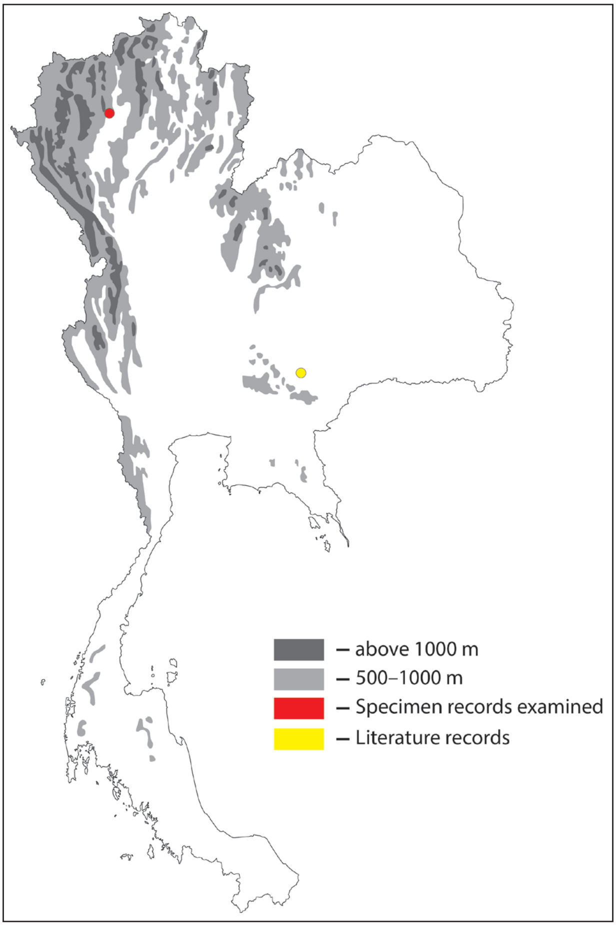


Figure 17. Thai distribution of *Megachile* (*Callomegachile*) *odontophora*.

Diagnosis. Female superficially resembles *M.* (*Callomegachile*) *atratiformis* (Meade-Waldo, 1914) in overall appearance and size: large body size (20–24 mm); black body covered with black hairs throughout; mesoscutum with weak transverse wrinkle pattern on disc, also posteriorly with weakly transverse wrinkle pattern; yellow wings (except apical margin of clypeus with median tubercle (Fig. 18e); mandibles four teeth with three stout apical teeth at apex and small tooth basally; labrum oblong with pointed apical margin and two lateral teeth (Fig. 18d)). Male can be easily recognized by clypeus covered with densely long hairs; forewings with black setae patch on medial cell; modified front tarsi (Engel and Gonzalez 2011).

Literature records. Myanmar. Thaungyin valley (Middle Tenasserim) (Engel and Gonzalez 2011); Thailand. Nakhon Ratchasima (Engel and Gonzalez 2011).

Material examined. Type material. Female paratype. Myanmar. “Middle Tenasserim; Thaungyin [= Moei River] Valley, 5/93, C.T. Bingham; Col. C.T. Bingham 96–30; Paratype, *Chalicodoma odontophorum* Michael S. Engel; NHMUK

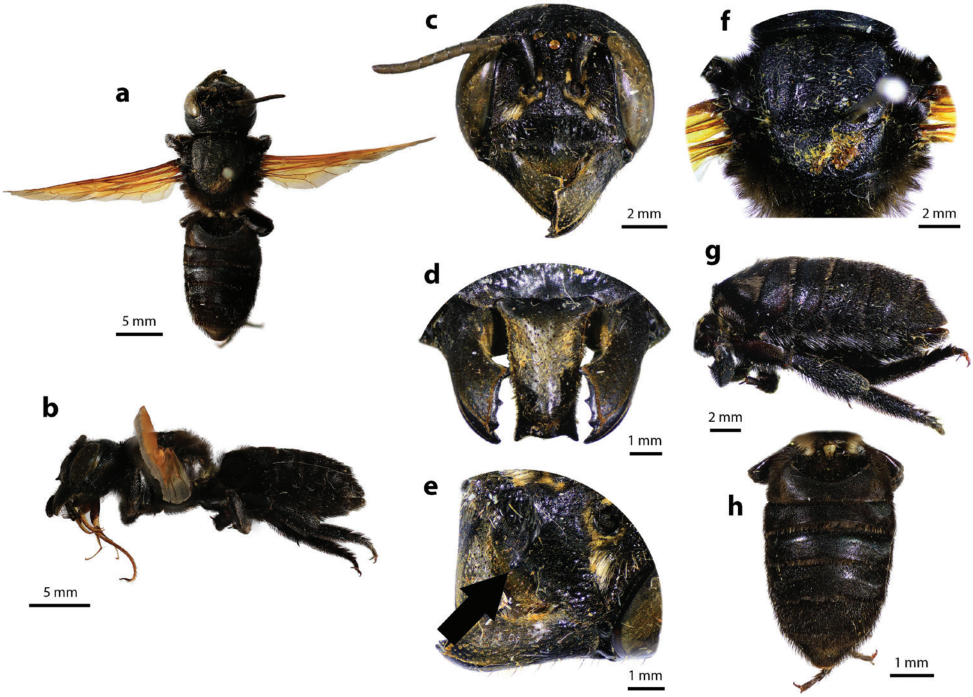


Figure 18. *Megachile* (*Callomegachile*) *odontophora* (Engel, 2011), female **a** dorsal view **b** lateral view **c** frontal view **d** frontal view of mandible and labrum **e** tubercle at clypeal margin (arrow) **f** dorsal view of mesoscutum **g** lateral view of metasoma and scopa **h** dorsal view of metasoma.

013380271"; Thailand. Chiang Mai Province: 1♀, 13-VII-2006, coll. M. Rungrote (leg. NC and NW).

Floral record. Engel and Gonzalez (2011) noted the species was captured on *Sindora siamensis* Teijsman & Miquel.

Megachile (*Callomegachile*) *ornata* Smith, 1853

Figs 19, 20

Megachile ornata Smith, 1853: 183; female syntype (NHMUK, examined) Indonesia.
Megachile miniata Bingham, 1896: 199.
Megachile ruficorbis Cockerell, 1927: 6.

Diagnosis. Female can be recognized by its black large body size (17–19 mm); T1–T4 covered with black hairs; T2 with small patch of brick-red hairs laterally; T5–T6 covered with pale light yellow hairs (Fig. 20a); mandible three teeth (Fig. 20b); second spine of pro- and mesotibiae bifurcate (Fig. 20c); metatibiae with spine at apex (Fig. 20d); brick-red scopa.

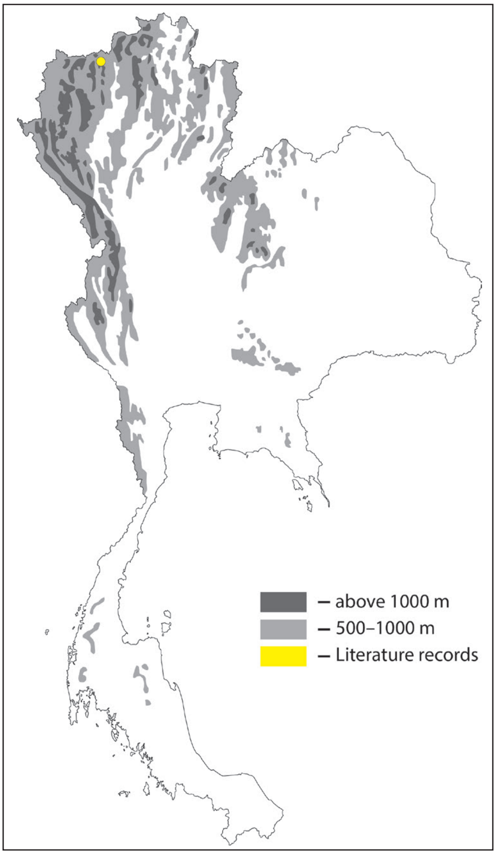


Figure 19. Thai distribution of *Megachile* (*Callomegachile*) *ornata*.

Literature records. Brunei. (Ascher and Pickering 2020); India. (Tadauchi and Tasen 2009); Indonesia. Borneo, Sumatra: including Deli (Kirby 1894; Meade-Waldo 1912; Tadauchi and Tasen 2009; Ascher et al. 2016); Malaysia. Kuala Lumpur, Negeri Sembilan, Pahang, Sabah, Sarawak, Selangor (Cockerell 1927a; Ascher et al. 2016; Ascher and Pickering 2020); Myanmar. (Cockerell 1927a); Nepal. (Tadauchi and Tasen 2009; Ascher and Pickering 2020); Singapore. (Ascher et al. 2016); Thailand. Chiang Mai (Tadauchi and Tasen 2009).

Material examined. Female syntype. Indonesia. "56 43; Locality unknown, pre-1853, Exchanged unit, Mr. Baly, B.M. 1856–43; Syntype, female, *Megachile ornata* Smith, F., 1853:183, det. D. Notton 2018 (ICZN Rec. 73F); B.M. TYPE HYM. 17a. 3215; NHMUK 013379840".

Comments. Trunz et al. 2016's phylogenetic analysis suggested *M. ornata* belongs to a distinct lineage of *Callomegachile* sensu lato.

Floral records. *Megachile* (*Callomegachile*) *ornata* was captured on *Grammatophyllum speciosum* Blume (Ascher et al. 2016).

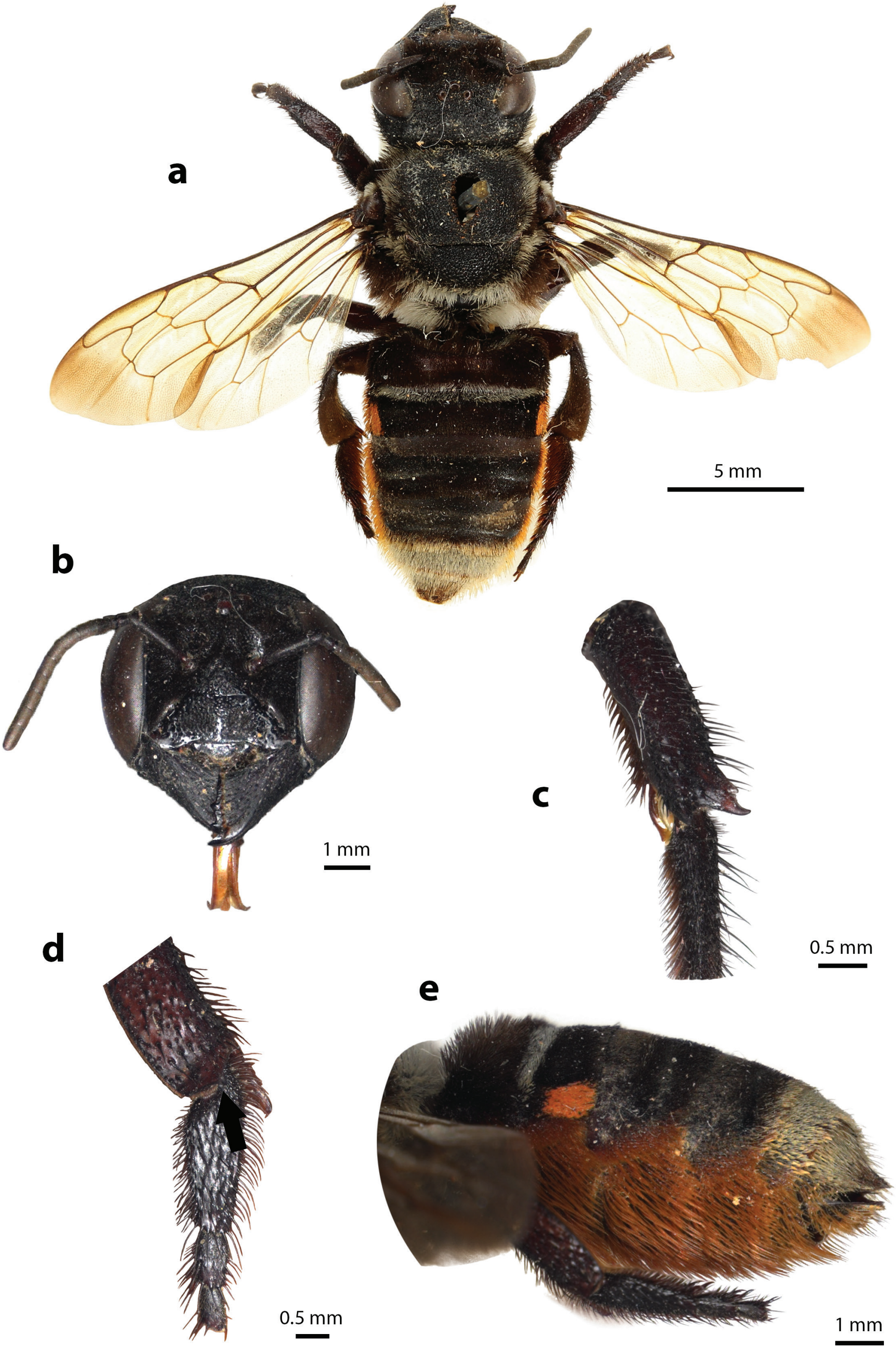


Figure 20. *Megachile* (*Callomegachile*) *ornata* Smith, 1853, female (NHMUK 013379840) **a** dorsal view **b** frontal view **c** spine at front tibia **d** spine at hind tibia **e** lateral view of mesosoma and scopa.

Megachile* (*Callomegachile*) *parornata **Chatthanabun, Warrit and Ascher, nom. nov.**

Figs 21, 22

Megachile gigas Wu, 2005: 159; preoccupied (junior primary homonym, not *Megachile gigas* Schrottky, 1908, Brazil). Female holotype (erroneously described as male) (IZB, examined) Xiaomengyang, Xishuangbanna, Yunnan, China.

Diagnosis. Female superficially resembles *M.* (*Callomegachile.*) *ornata* Smith, 1853 except T1 covered with black hairs; T2–T5 covered with brick-red hairs; T6 covered with pale yellow hairs (Fig. 22g, h); scopa brick-red.

Redescription. *Female.* Length. Total body length 20.00–21.50; wingspan 35.55–38.24; forewing 14.84–16.13. Structure and color. Head black; ventral margin of paraocular area with brown hairs; clypeus trapezoid with rough surface and sparse punctures;

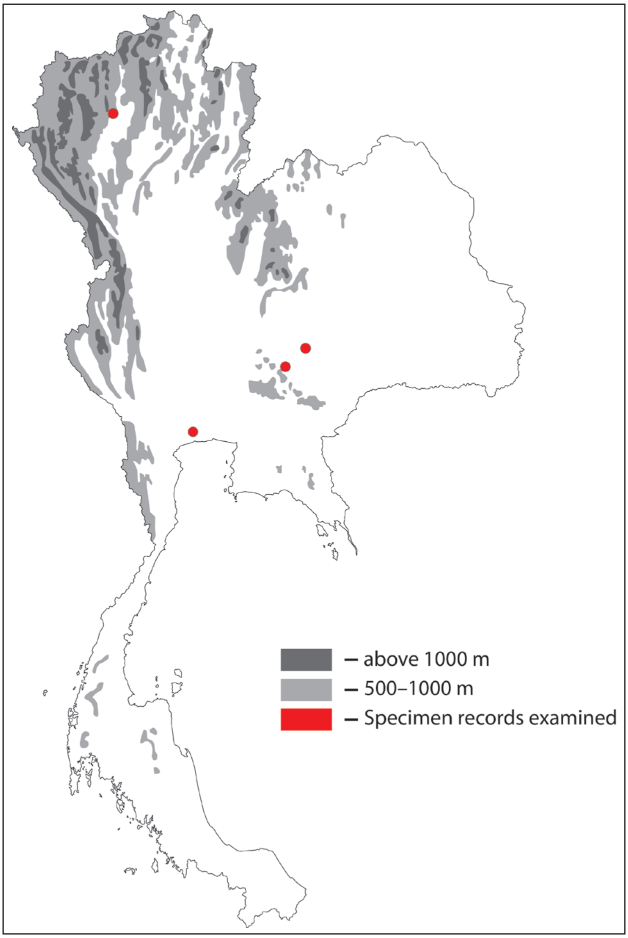


Figure 21. Thai distribution of *Megachile* (*Callomegachile*) *parornata*.

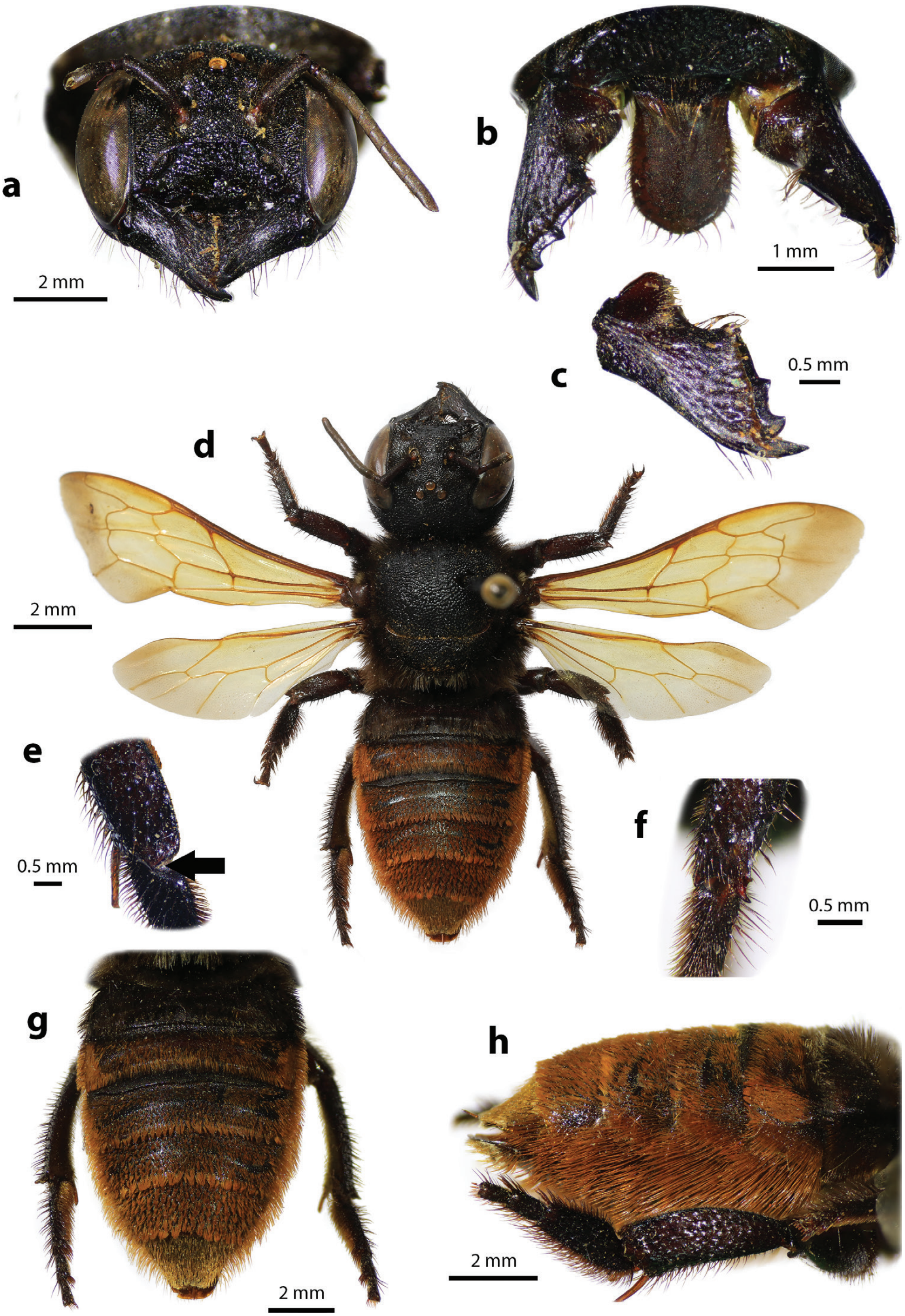


Figure 22. *Megachile* (*Callomegachile*) *parornata* Chatthanabun, Warrit and Ascher, nom. nov., female **a** frontal view **b** labrum and mandibles **c** mandible **d** dorsal view **e** spine on hind tibia **f** spine on front tibia **g** dorsal view of metasoma **h** lateral view of metasoma and scopa.

supraclypeal area convex and subtriangular with rough surface and sparse punctures; mandible with three teeth, without cutting edge; outer surface of mandible minutely roughened with long brown hairs; labrum length twice as long as wide with round apex, surface minutely convex and rough with erect long brown hairs at apex; gena with sparse punctures; bare vertex with sparse punctures, ID shorter than OD, ID/OD = 0.28 ± 0.01; antennae with eleven flagella, first flagellomere wider than long, shorter than the second; body parallel-sided; scutum and scutellum hairless except anterior margin of scutum with brown hairs; lower part of metathorax with white hairs; scutum and lower part of mesepisternum with coarsely striate puncture pattern; procoxa covers with brown hairs; pro- and mesotibiae with two apical spines, mesotibial spine bifurcate; apex of metatibiae with spine at apex; pro- and mesotarsus with short brown hairs; metatarsus with dense short fulvous hairs inner side and short brown hairs outer side; forewing length yellow hyaline with yellowish-brown vein; T1 covers with black hairs; T2–T5 cover with brick-red hairs; T6 covers with pale yellow hairs, round apex; scopa fulvous-red.

Male. Unknown.

Literature records. China. Yunnan (Wu 2005); Vietnam. Hoa-Binh, Vinh-Quang (Wu 2005).

Material examined. Female holotype (erroneously described as male). China. "Yunnan, Xishuangbanna, Xiaomengyang (22°N, 100.8°E), 850 m; 1957. IX. 6, collected by Zang Ling-Chao; Holotype; female, *Megachile* (*Callomegachile*) *gigas* Wu, 2005; det. Y. R. Wu; IOZ(E) 210406"; Thailand (new record). Bangkok Province: 1♀, Dusit, 8-IX-1968, coll. Patchanee (leg. NC and NW). Chiang Mai Province: 1♀, Meuang district, Faculty of Agriculture, 4-VIII-1981, coll. Vijit (leg. NC and NW); 1♀, Meuang district, Chiang Mai University, 6-VIII-1981, coll. Sumrid (leg. NC and NW). Nakhon Ratchasima Province: 1♀, 30-VI-1962, coll. unknown (leg. NC and NW); 1♀, Faculty of Forestry, 10-VII-1968, coll. K. Vajropala (leg. NC and NW).

Etymology. The species name refers to the close resemblance to *M. ornata*.

Remarks. The wide published distribution of *M. ornata* from Nepal to the Indonesian Archipelago raises a question regarding whether this is a single species or a species complex. Since the type material of *M. ornata* was collected from Sumatra, Indonesia, specimens reported under this name from other mainland Asian countries should be re-examined. Furthermore, whereas no additional specimens of *M. parornata* were collected and deposited in NHMCU-BSRU and CMU since 1968, Tadauchi and Tasen (2009) recorded one specimens of *M. ornata* from Chiang Mai province (not seen by us). Until, male specimens of both *M. parornata* and *M. ornata* are collected and studied, we are proposing *M. parornata* to be a separate species from *M. ornata*.

***Megachile* (*Callomegachile*) *tuberculata* Smith, 1858**

Figs 23, 24

Megachile tuberculata Smith, 1858: 46. Female syntype (NHMUK, examined) Borneo, Malaysia.

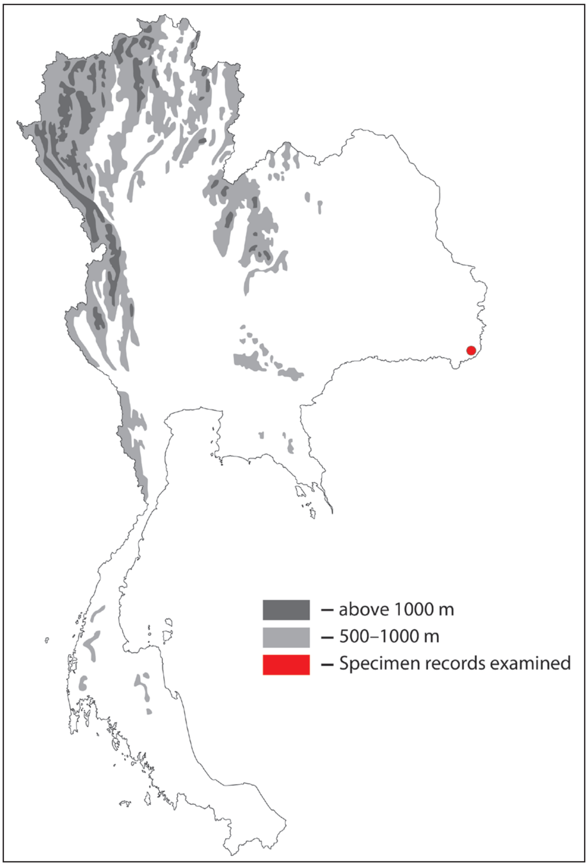


Figure 23. Thai distribution of *Megachile* (*Callomegachile*) *tuberculata*.

Megachile longipalpis Radoszkowski, 1882: 78 [doubtful synonymy].
Chalicodoma (*Eumegachilana*) *tuberculatum*: Michener, 1965: 192.

Diagnosis. Female can be recognized by its large body size (21–24 mm); black body covered with black hairs throughout (Fig. 24a); base of clypeus with large protruding tubercle; mandibles elongate with three teeth and small tubercle at base (Fig. 24c); yellow wings; black scopa.

Literature records. India. Sikkim (Ascher et al. 2016); Indonesia. Java, Sumatera Barat (Ascher et al. 2016); Myanmar. Mergui Archipelago, Bago Yoma (Bingham 1890, 1897); Malaysia. Johor, Negeri Sembilan, Pahang, Perak, Sabah, Sarawak, Selangor, Terengganu, (Smith 1858; Bingham 1890, 1897; Dover 1929; Ascher et al. 2016); Philippines. (The Philippines record based on the type of *M. longipalpis* Radoszkowski, 1882 is considerably outside the verified range for this species as presently understood, so this synonymy should be re-examined); Singapore. (Bingham 1897; Ascher et al. 2016).

Material examined. Female syntype. Malaysia. "Type; B.M. TYPE HYM. 17a. 2840; SAR.; *Megachile tuberculata* Sm. 1857 not 1879; NHMUK 013379846"; Thai-

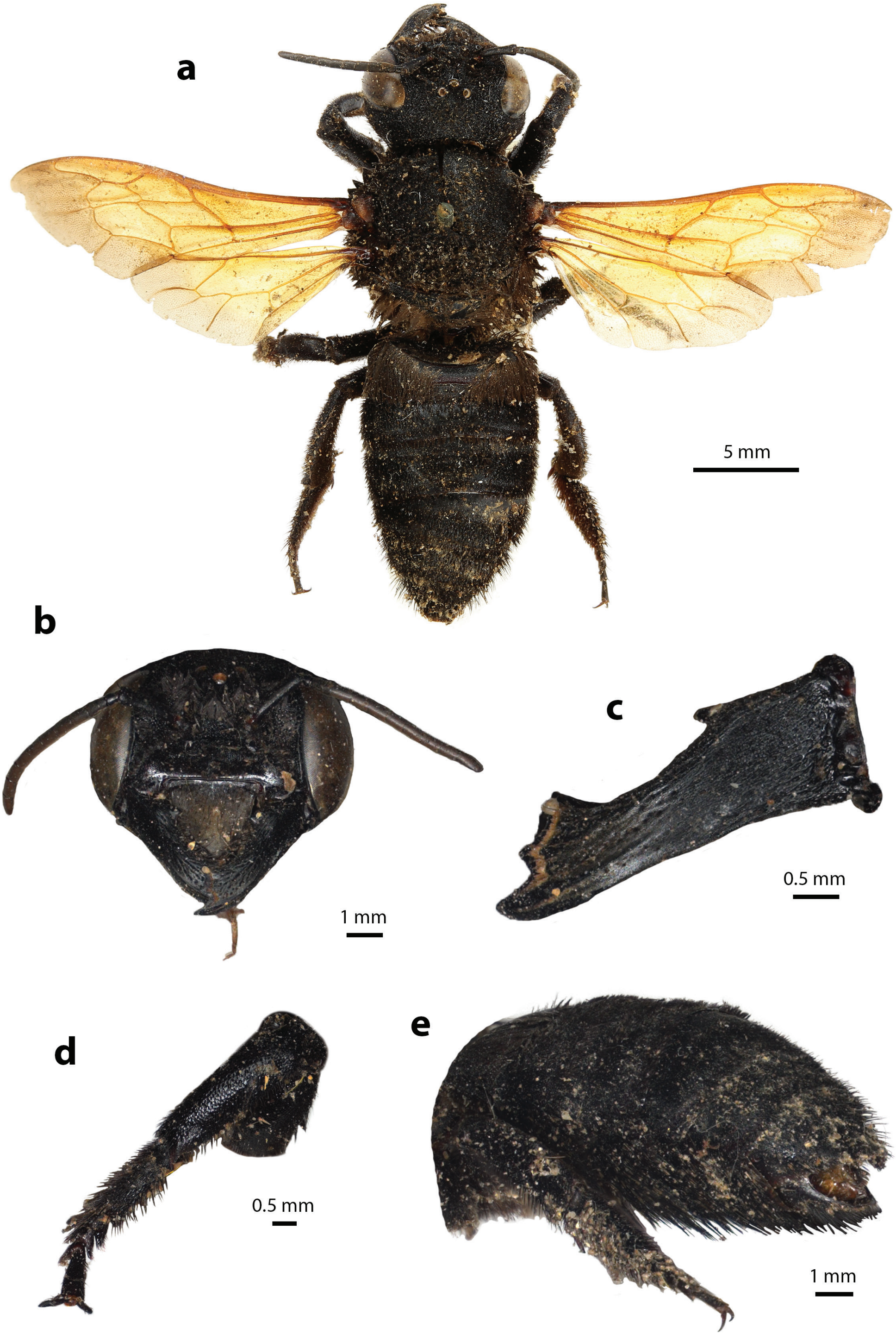


Figure 24. *Megachile* (*Callomegachile*) *tuberculata* Smith, 1858, female (NHMUK 013379846) **a** dorsal view **b** frontal view **c** lateral view of mandible **d** front leg **e** lateral view of metasoma and scopa.

LAND (new record). Ubon Ratchathani Province: 1♀, Na Chaluai district, Phu Jong Nayoy Nat. P., Phalan Pa Chad, 14°26'7.8066"N, 105°15'34.7394"E, Alt. 247.17 m, 6-XI-2019, coll. Traiyasut et al. (leg. NC and NW).

Floral records. *Megachile* (*Callomegachile*) *tuberculata* was photographed visiting and collecting pollens from *Psophocarpus tetragonolobus* (L.) D.C. (Soh et al. 2017), also captured on *Grammatophyllum speciosum* Blume (Ascher et al. 2016).

Comments. The single Thai record is from far to the northeast of the Sundaic Region.

Megachile (*Callomegachile*) *umbripennis* Smith, 1853

Figs 25–29

Megachile umbripennis Smith, 1853: 175. Female syntype (NHMUK, examined) Nepal [as Nepaul].

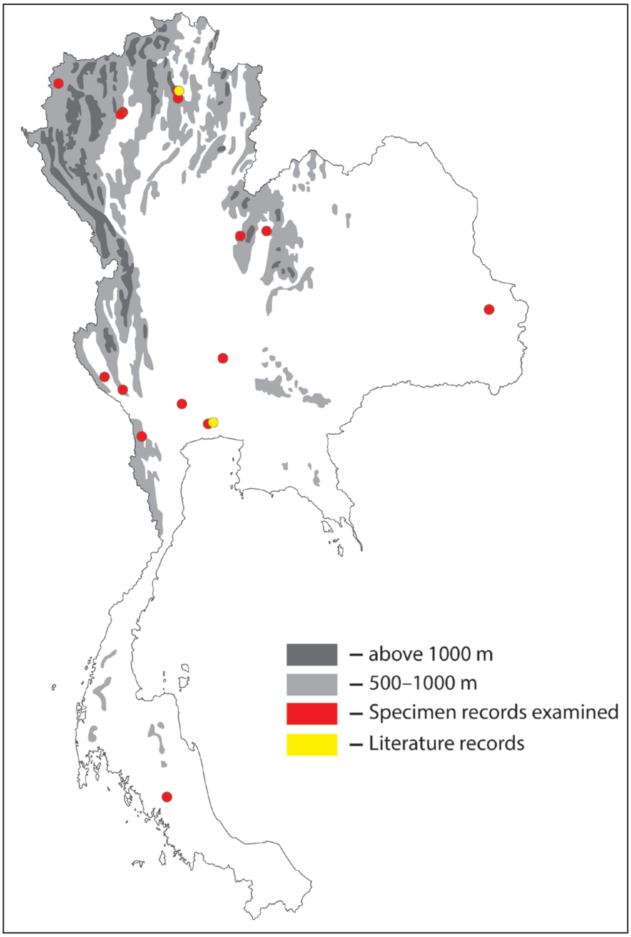


Figure 25. Thai distribution of *Megachile* (*Callomegachile*) *umbripennis*.

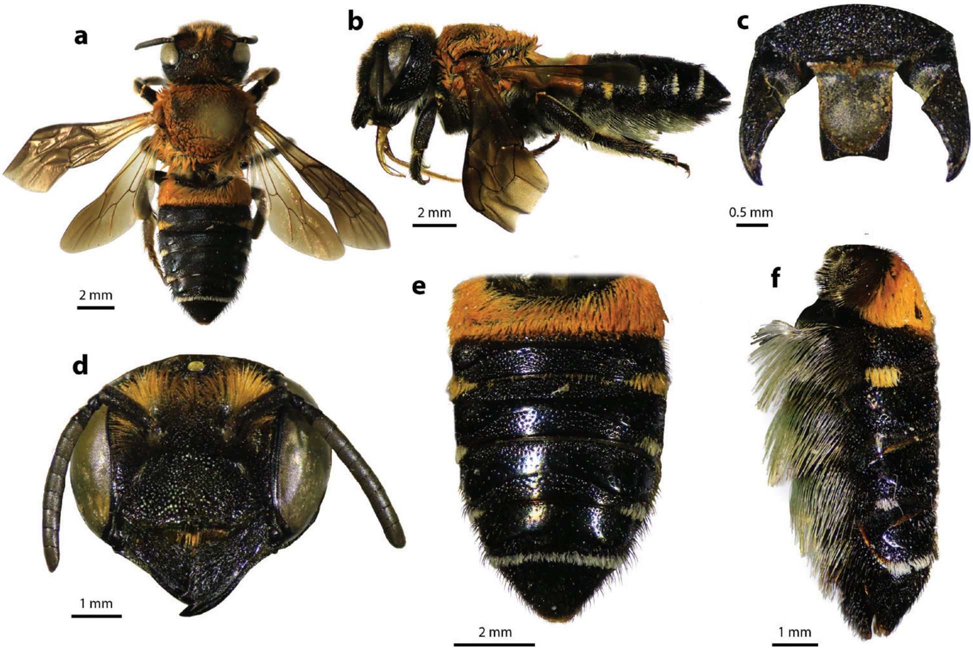


Figure 26. *Megachile* (*Callomegachile*) *umbripennis* Smith, 1853, female **a** dorsal view **b** lateral view **c** frontal view of mandible and labrum **d** frontal view **e** dorsal view of metasoma **f** lateral view of metasoma and scopa.

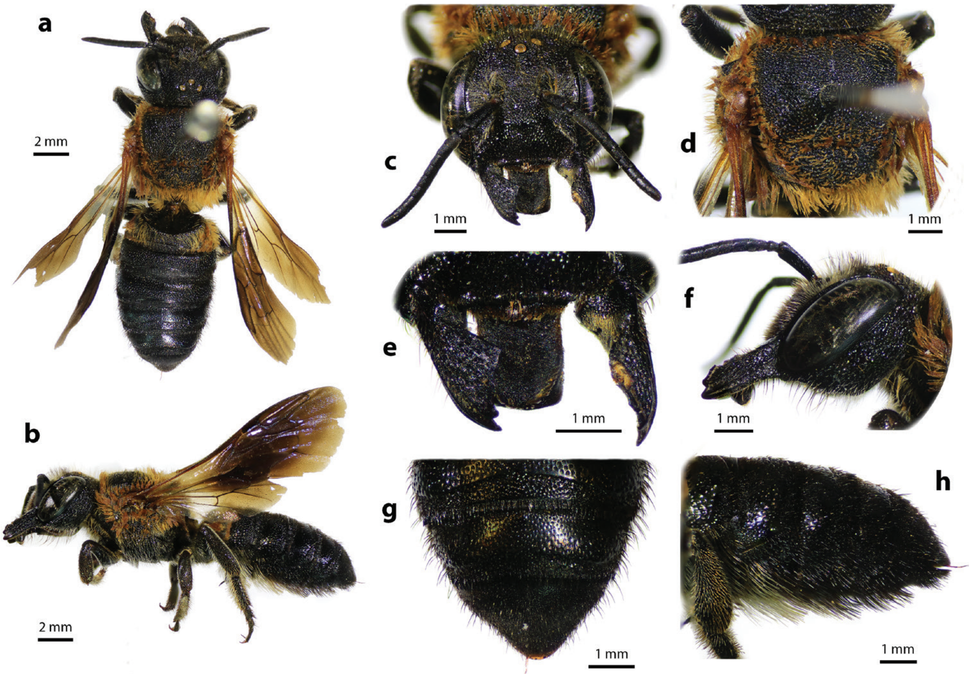


Figure 27. *Megachile* (*Callomegachile*) *umbripennis* Smith, 1853 (BSRU AA-4620), female **a** dorsal view **b** lateral view **c** frontal view **d** dorsal view of mesoscutum **e** clypeus and mandible **f** lateral view of mandible **g** T6 **h** lateral view of metasoma showing scopa.

Megachile schauinslandi Alfken, 1898: 340.
Megachile domesticum Perkins, 1899: 114, nomen nudum.
Megachile umbripennis var *atriventris* Friese, 1903: 357.
Megachile aureobasis Cockerell, 1919: 198.
Megachile (*Eumegachile*) *umbripennis*: Krombein, 1950: 125.
Chalicodoma (*Callomegachile*) *umbripennis*: Michener, 1965: 191.

Diagnosis. Female superficially resembles leafcutter bee *M.* (*Aethomegachile*) *laticeps* Smith, 1853, in terms of its overall appearance and size: mesosoma and propodeal triangle with fulvous hairs, except vertex and disc area of mesoscutum covered with fulvous hairs (Fig. 26a); T2–T4 with tuft fulvous hairs laterally; T5 with white hair band (Fig. 26e). Male is similar to female except apical margin of clypeus covered with hairs; mandibles three teeth (Fig. 28b, c).

Literature records. American Samoa; China. Aitutaki, Canton, Fujian, Guangdong, Sichuan, Yunnan (Ascher et al. 2016); Fiji. (Davies et al. 2013; Ascher et al. 2016); French Polynesia. Mo'orea, Society Islands, Tahiti (Ascher et al. 2016); Hong Kong. (Ascher et al. 2016); India. Chandigarh, Haryana, Sikkim, Tamil Nadu (Bingham 1897; Ascher et al. 2016); Laos. Vientiane (Ascher et al. 2016); Malaysia. Kelantan, Kuala Lumpur, Pahang, Sarawak, Selangor (Smith 1858; Ascher et al. 2016); Mariana Islands. San Vicente; Myanmar. Tenasserim (Bingham 1897; Ascher et al. 2016); Nepal. (Smith 1853, 1858; Bingham 1897; Ascher et al. 2016); New Caledonia. (Ascher et al. 2016); Northern Mariana Islands. (Ascher et al. 2016); Singapore. (Ascher et al. 2016); Sri Lanka. Matale (Ascher et al. 2016); Cook Islands. Aitutaki, Rarotonga, Mangaia (Kuhlmann 2006; Ascher et al. 2016); Tonga. (Ascher et al. 2016); USA. Florida, Hawaii, Kauai, Midway, Maui, Molokai, Oahu (Ascher et al. 2016); Vietnam. Ha Giang (Ascher and Pickering 2020).

Material examined. Female syntype. Nepal. "Type; B.M. TYPE HYM. 17. a. 2160; Hardwicke Bequest; Nepal [as Nepaul]; umbripennis, Type, Sm.; *Megachile umbripennis* TYPE. Sm.; NHMUK 013380267"; Thailand. Chiang Mai Province: 1♀, 16♂, Mueang district, Mae Hia subdistrict, Mae Hia Agricultural Research, Demonstrative and Training Center, 18°45'51.1272"N, 98°55'39.6192"E, Alt. 232 m, 19-VII-2015, coll. Warrit et al. (leg. NC and NW). Kanchanaburi Province: 1♀, 19♂, Sai Yok district, Wang Krachae subdistrict, 14°9'56.7678"N, 98°55'39.6192"E, Alt. 232 m, 24-VI-2016, coll. Warrit et al. (leg. NC and NW); 8♂, Sai Yok district, Wang Krachae subdistrict, 14°11'6.5724"N, 99°3'6.9258"E, Alt. 102.30 m, 24-VI-2016, coll. Warrit et al. (leg. NC and NW); 1♀, Sai Yok district, Sai Yok Yai National Park, 14°27'16.4118"N, 98°51'40.4928"E, Alt. 49.17 m, 24-VI-2016, coll. Warrit et al. (leg. NC and NW). Mae Hong Son Province: 1♀, Pang Ung, Pang Tong Under Royal Forest Park 2, 19°29'58.3008"N, 97°54'42.1014"E, Alt. 1,164 m, 10-XII-2015, coll. Warrit et al. (leg. NC and NW). Nakhon Pathom Province: 1♀, Kamphaeng Saen district, Kamphaeng Saen subdistrict, Kasetsart University, Kamphaeng Saen Campus, 22-II-2000, coll. Sokroh (leg. NC and NW); 1♀, Kamphaeng Saen district, Kamphaeng Saen subdistrict, Kasetsart University, Kamphaeng Saen Campus, 22-II-2003, coll. Kittisak (leg. NC and NW); 2♂, Kamphaeng Saen district, 13°44'58.3908"N, 99°52'33.1242"E, Alt. 14 m, 10-VII-2015, coll. Warrit et al. (leg. NC

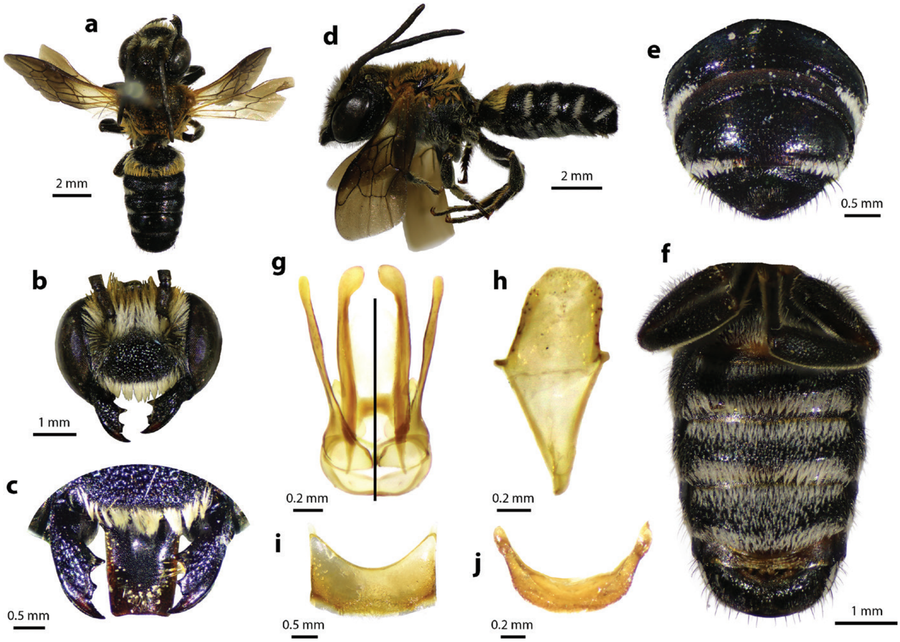
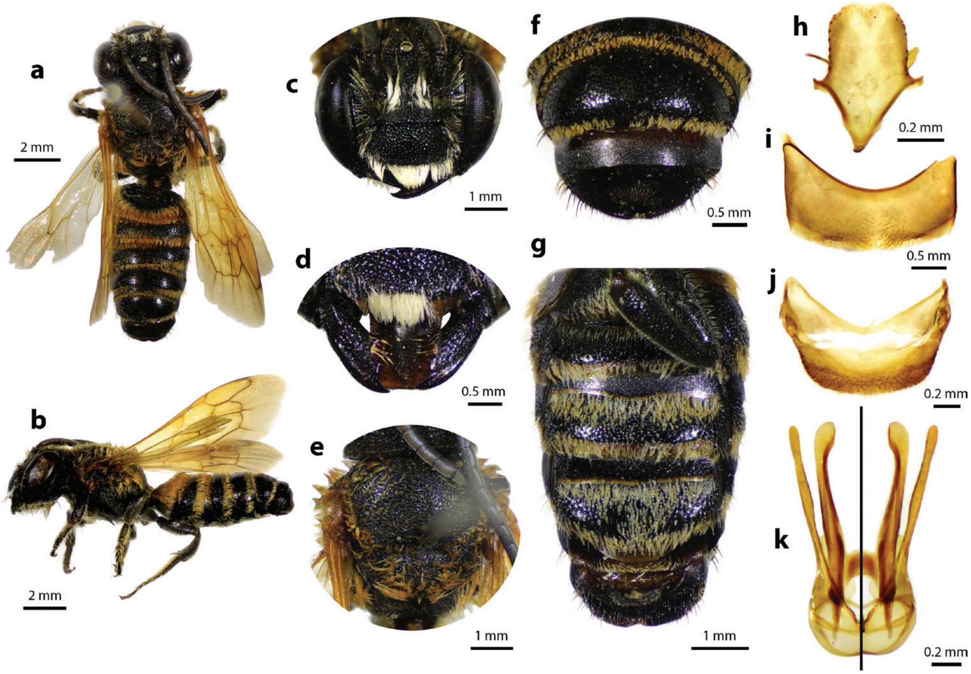


Figure 28. *Megachile* (*Callomegachile*) *umbripennis* Smith, 1853, male **a** dorsal view **b** frontal view **c** frontal view of mandible and labrum **d** lateral view **e** frontal view of T7 **f** ventral view of metasomal sterna **g** dorsal (left) and ventral (right) views of penis **h** dorsal view of S8 **i** S5 **j** T7.

and NW). Phayao Province: 1♀, 4♂, Mueang district, Maeka subdistrict, Phayao University, 01-VI-2012, coll. Warrit et al. (leg. NC and NW); 1♀, 28-I-2014, coll. S. Yutham (leg. NC and NW). Phetchabun Province: 2♂, Lomsak district, Bungkla subdistrict, 19-X-2009, coll. K. Attasopa & P. Phukphume (leg. NC and NW); 1♀, 2♂, Lomsak district, Bungkla subdistrict, 20-X-2009, coll. K. Attasopa & P. Phukphume (leg. NC and NW); 1♀, Lomsak district, Bungkla subdistrict, 21-X-2009, coll. K. Attasopa & P. Phukphume (leg. NC and NW). Phitsanulok Province: 1♀, Phuhinrongkla Nat. P., Lan hin pum, 16°59'49.8008"N, 101°00'40.6772"E, Alt. 1303 m, VI. 17. 2017., Aerial net, coll N. Warrit et al. (leg. NC and NW). Ratchaburi Province: 1♀, Suan Pheung district, 25-V-2012, coll. N. Warrit (leg. NC and NW). Samut Sakhon Province: 1♀, Banpaew district, 19-VIII-2014, coll. P. Tangtorwongsakul (leg. NC and NW). Suphan Buri Province: 1♂, 9-X-2012, coll. Veerawan (leg. NC and NW). Trang Province: 1♂, Na Yong district, 7°33'8.0892"N, 99°46'33.6072"E, Alt. 24 m, 11-VI-2015, coll. Warrit et al. (leg. NC and NW). Ubon Ratchathani Province: 1♂, Trakan Phuetphon district, Sarin lake view village, 03-VIII-2014, coll. N. Chatthanabun (leg. NC and NW).

Comments. There are some suspect specimens that show variation in both sexes of *M. umbripennis*. One female collected from Phitsanulok province (BSRU AA-4620) shows the following variations: lack of fulvous hairs on disc area of mesoscutum and lack of white hairs on T2–T5 (Fig. 27a–h). Males collected from Chiang Mai province (BSRU AA-3654, BSRU AA-3662) and Suphan Buri province (KKIC-02) have fulvous hairs on T2–T5 instead of white hairs (Fig. 29a–k).



Figures 29. *Megachile* (*Callomegachile*) *umbripennis* Smith, 1853 (BSRU AA-3654, BSRU AA-3662 and KKIC-02), male **a** dorsal view **b** lateral view **c** frontal view **d** clypeus and mandible **e** dorsal view of mesoscutum **f** T6 **g** ventral view of metasomal sterna showing scopa dorsal view of metasomal sterna **h** dorsal view of S8 **i** S5 **j** T7 **k** dorsal (left) and ventral (right) views of penis.

***Megachile* (*Callomegachile*) *chiangmaiensis* Chatthanabun & Warrit, sp. nov.**

http://zoobank.org/037E992C-1DB2-4BA4-B5EB-6684A3F4374F
Figs 30, 31

Diagnosis. The species superficially resembles *M. disjuncta* (Fabricius, 1781) in terms of its overall appearance and size: white tuft of hairs on scutellum, propodeum, and first few segments of metasomal terga; however, the prominent apical half-circular impression of clypeus with strong median carina (Fig. 31e) differentiates *M. chiangmaiensis* sp. nov. from the former. Clypeal impression smooth with dense dark hairs at apex. Such a clypeal impression was also present in another species of *Callomegachile*, *M. ramakrishnae* (Cockerell, 1919), a rare bee collected in Tamil Nadu, India, although the impression in *M. ramakrishnae* is more or less shallower and the pattern of the mesosoma and white hairs band on T2–T3 are absent. The apex of the labrum is strongly pointed medially with two lateral teeth (Fig. 31f).

Description. ***Female.*** Length. Total body length 12.57–13.64; wingspan 18.60–22.56; fore wing 9.66 Structure and color. Head black; paraocular area with dense black hairs; clypeus with prominent apical half-circular impression with strong median carina; clypeal impression smooth with dense dark hairs at apex; smooth area of subtriangular supraclypeal with sparse punctures; mandible stout and elongate with-

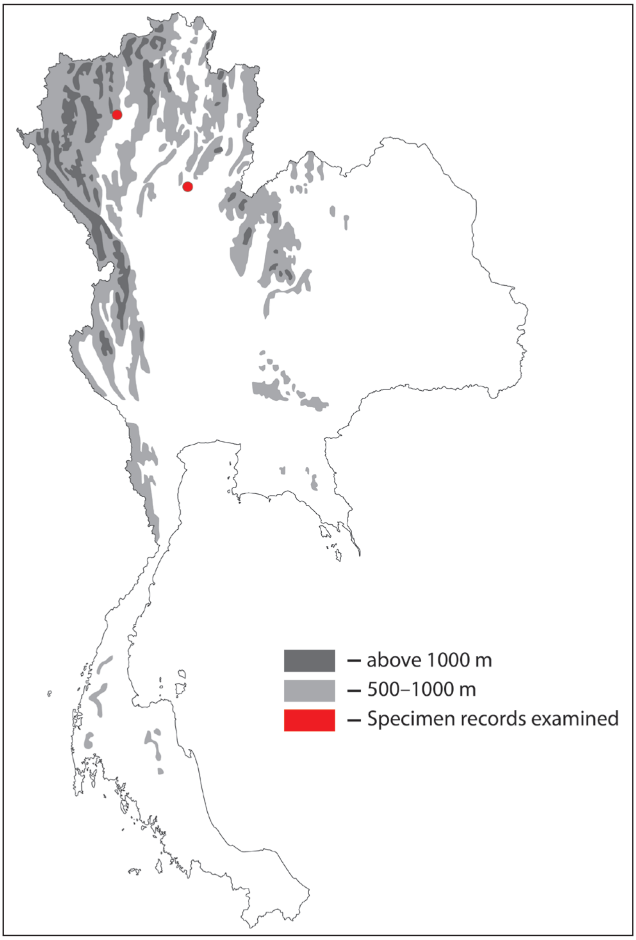


Figure 30. Thai distribution of *Megachile* (*Callomegachile*) *chiangmaiensis* new species.

out cutting edge, mandible with three apical teeth, outer surface minutely roughened with long brown hairs; surface of labrum minutely roughened, apex of labrum medially strongly pointed with two lateral teeth; gena with sparse punctures; sparse punctures on vertex with ID shorter than OD, ID/OD = 0.55 ± 0.06; antennae with ten flagella, first flagellomere wider than long and shorter than the second; body parallel-sided; mesoscutum and lower part of mesepisternum with coarsely striate puncture pattern; mesoscutellum and propodeum with tuft of white hairs; procoxa with small ridge, covered with sparse brown hairs; pro- and mesotibiae with two spines at apices; apex of metatibiae truncate; pro-, meso- and metatarsi with dense brown short hairs; hyaline wings with smoky color at apex and dark brown veins; T1 and pregradular area of T2 covered with tuft of white hairs; T2–T5 with dense punctures on margin and short black hairs on each side; T6 covered with short black hairs, apex round shape; scopa black.

Male. Unknown.

Distribution. Thailand. Chiang Mai and Uttaradit.

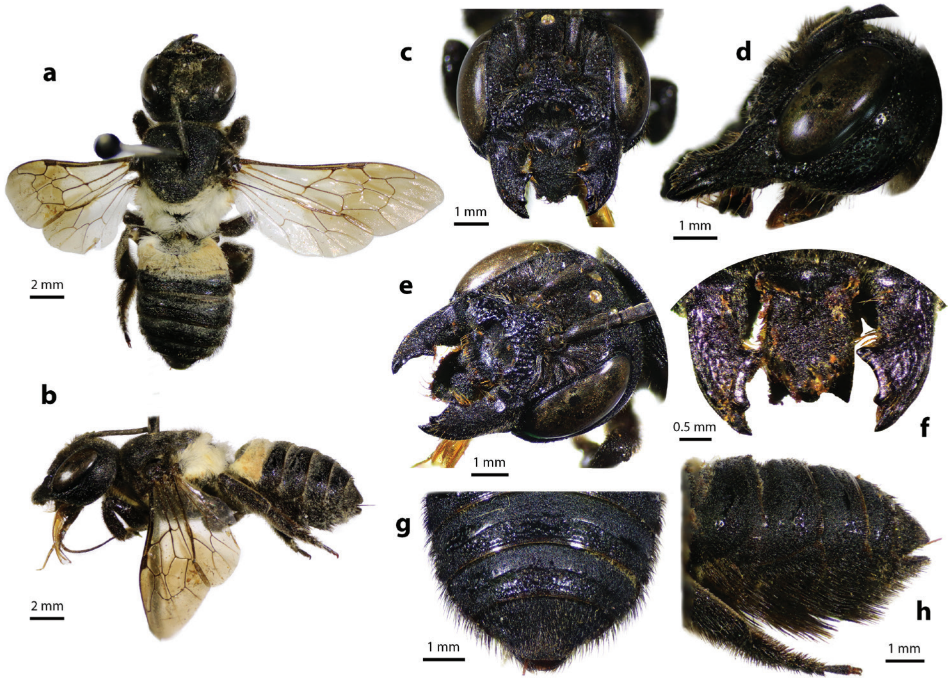


Figure 31. *Megachile* (*Callomegachile*) *chiangmaiensis* Chatthanabun and Warrit, sp. nov., holotype, female **a** dorsal view **b** lateral view **c** frontal view **d** lateral view of mandible **e** oblique view of clypeus **f** clypeus and mandible **g** T6 **h** lateral view of metasoma showing scopa.

Material examined. Female holotype. THAILAND. "เชียงใหม่, 11 ก.ย. 56, วิมลชัย [Chiang Mai, 11 September 2013, coll. Vimolchai]" (KKIC-01); Female paratypes. THAILAND. "Uttaradit, 8 April 1961, coll. unknown" (3♀, DNP-0002, DNP-0003, DNP-0004).

Etymology. The new species is named after the type locality.

Remarks. *Megachile chiangmaiensis* sp. nov. can be found in the same province as the morphologically similar congeneric species, *M. disjuncta*, although the latter are abundantly collected throughout Thailand. The biology of *M. chiangmaiensis* is unknown.

Megachile (*Carinula*) Michener, McGinley, & Danforth, 1994

Chalicodoma (*Carinella*) Pasteels, 1965: 447. Type species: *Megachile torrida* Smith, 1853, by original designation.

Megachile (*Carinula*) Michener, McGinley, & Danforth, 1994: 174, replacement for *Carinella* Pasteels, 1965. Type species: *Megachile torrida* Smith, 1853, autobasic.

Diagnosis. Body size median to small. Female mandible four to five teeth. Clypeus with median carina. Clypeal margin crenulate with five teeth. In males, coxal spine absent, front tarsi simple and carina of T6 extremely reduced.

Comments. *Carinula* is superficially similar to *Callomegachile*, especially striated punctures on mesoscutum and lower part of mesepisternum. Female of *Carinula* can be recognized by the presence of crenulate clypeal margin, whereas male can be recognized by reduced carina on T6.

Megachile (*Carinula*) *stulta* Bingham, 1897

Figs 32, 33

Megachile stulta Bingham, 1897: 476; Female syntype (NHMUK, examined) Sikkim, India.

Diagnosis. Female can be recognized by its medium to small body size (7.83–10.39 mm); rough clypeus with median carina, apical margin crenulate (Fig. 33b); mandibles four teeth with two stout apical teeth at apex and two small teeth basally; labrum rectangular (Fig. 33c); metasoma covered with ferruginous hairs (Fig. 33e); scopa ferruginous except white basal area (Fig. 33f).

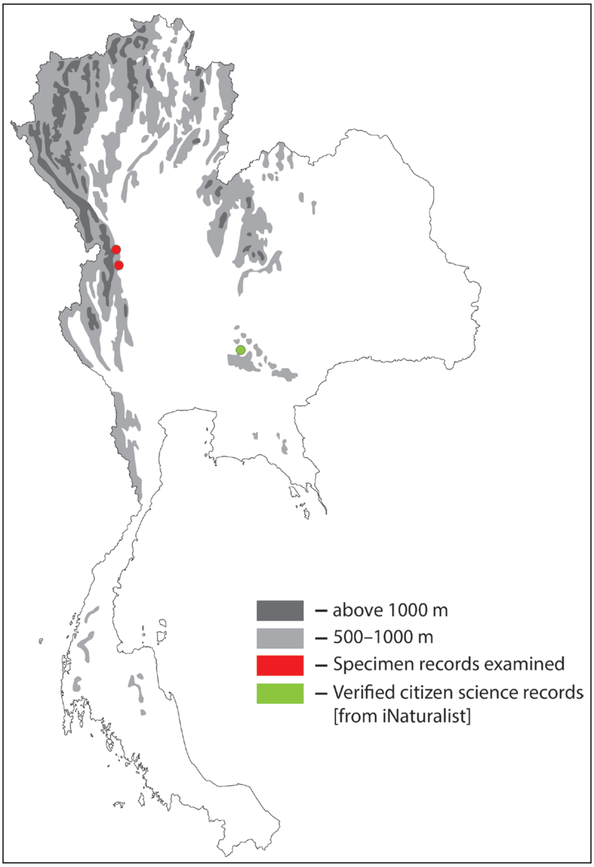


Figure 32. Thai distribution of *Megachile* (*Carinula*) *stulta*.

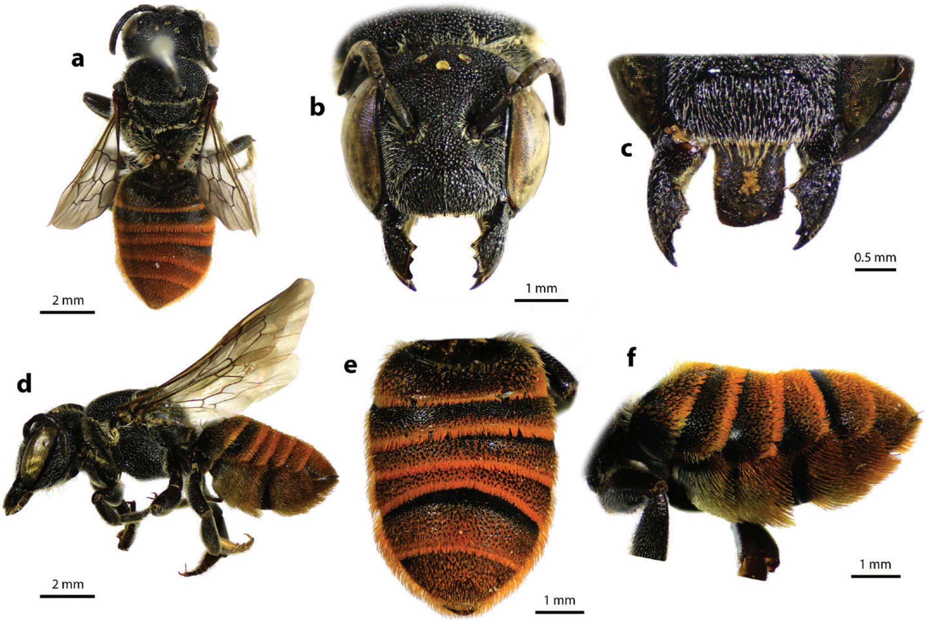


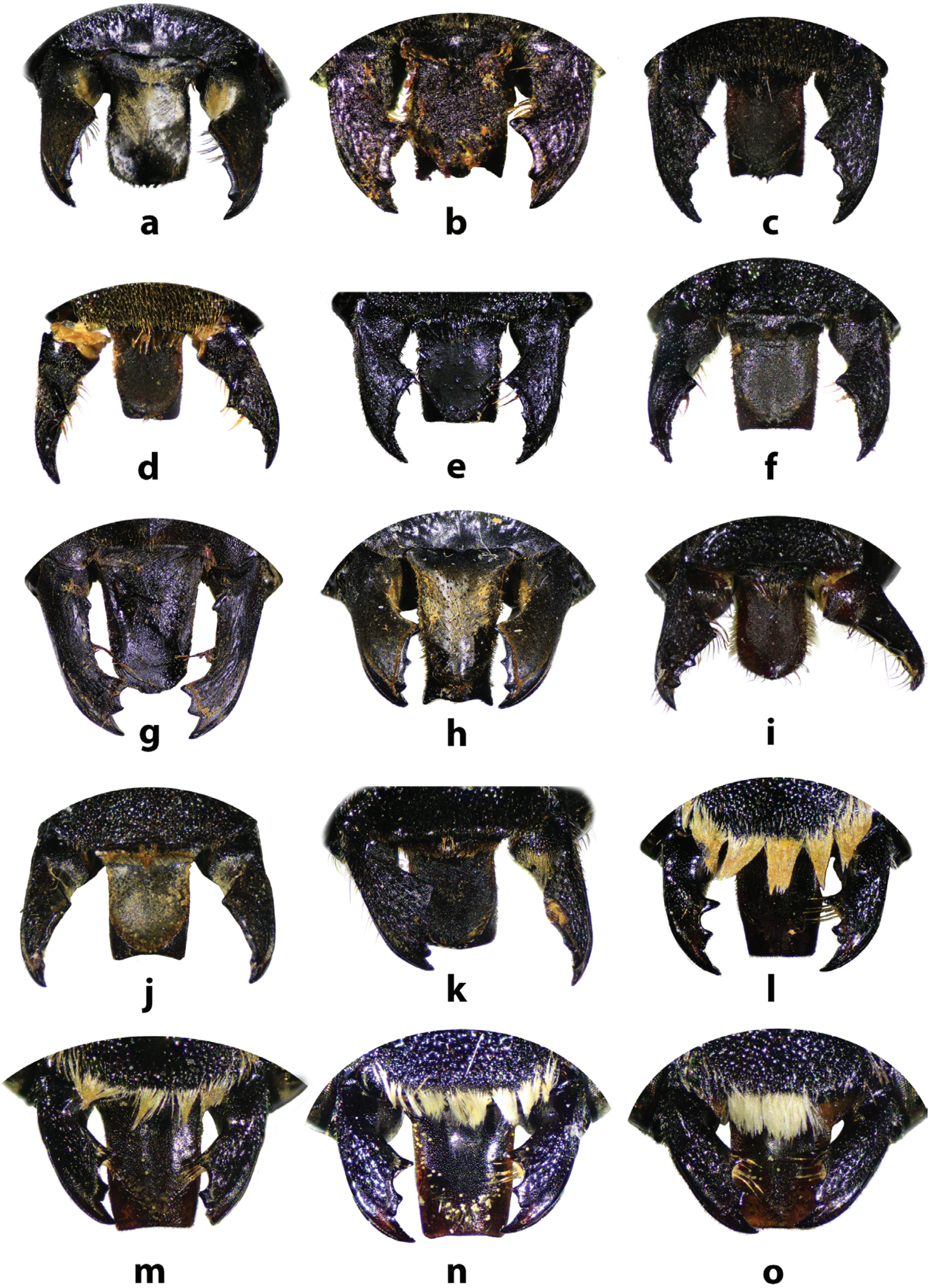
Figure 33. *Megachile* (*Carinula*) *stulta* Bingham, 1897, female **a** dorsal view **b** frontal view **c** frontal view of mandible and labrum **d** lateral view **e** dorsal view of metasoma **f** lateral view of metasoma and scopa.

Literature records. India. Karnataka, Sikkim (Ascher et al. 2016; Ascher and Pickering 2020); Indonesia. Sumatra (Bingham 1897; Ascher and Pickering 2020); Malaysia. Kuala Lumpur, Selangor (Ascher et al. 2016; Ascher and Pickering 2020); Myanmar. Tenasserim (Bingham 1897); Singapore. (Ascher et al. 2016). In addition, six females of this species was among numerous megachilids photographed together at Hin Tung, Mueang District, Nakhon Nayok Province (iNaturalist) (credit: scottyastro 2015; shuanda 2019).

Material examined. Female syntype. India. "Type; B.M. TYPE. 17.a.2161b; *Megachile stulta* Bingh, female, Type.; SIKKIM, Rungjit Valley, 1000 ft., 4.94, BINGHAM COLL., Col. C. T. Bingham 96–30; NHMUK 013380269"; Thailand. Kamphaeng Phet Province: 1♀, Khlong Lan, Khlong Lan waterfall, 08-IV-2014, coll. C. Wimolsuthikul & S. Wongvilas (leg. NC and NW); 36♀, Pang Sila Thong district, Mae Wong National Park, Kang Pha Khoi Nang, 07-VIII-2015, coll. N. Warrit et al. (leg. NC and NW).

Notes on *Callomegachile* from Thailand

Two of the most common species of *Megachile* (*Callomegachile*) sensu lato found in Thailand are *M.* (*Callomegachile*) *disjuncta* and *M.* (*Callomegachile*) *umbripennis*, and these are also the most common species of this group in Singapore (Ascher et al. 2016). *Megachile* (*Callomegachile*) *disjuncta* is usually collected from *Crotalaria juncea* L. across most of Thailand.



Figures 34. Distal edges of labrums and mandibles of some Thai *Callomegachile* (**a–k** females, **l–o** males) **a** *M.* (*Callomegachile*) *atratiformis* **b** *M.* (*Callomegachile*) *chiangmaiensis* sp. nov. **c, l** *M.* (*Callomegachile*) *disjuncta* **d, m** *M.* (*Callomegachile*) *faceta* **e** *M.* (*Callomegachile*) *fulvipennis* **f** *M.* (*Callomegachile*) *impressa* **g** *M.* (*Callomegachile*) *monticola* **h** *M.* (*Callomegachile*) *odontophora* **i** *M.* (*Callomegachile*) *parornata* **j, n** *M.* (*Callomegachile*) *umbripennis* **k** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-4620) **o** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-3654, BSRU AA-3662 and KKIC-02).

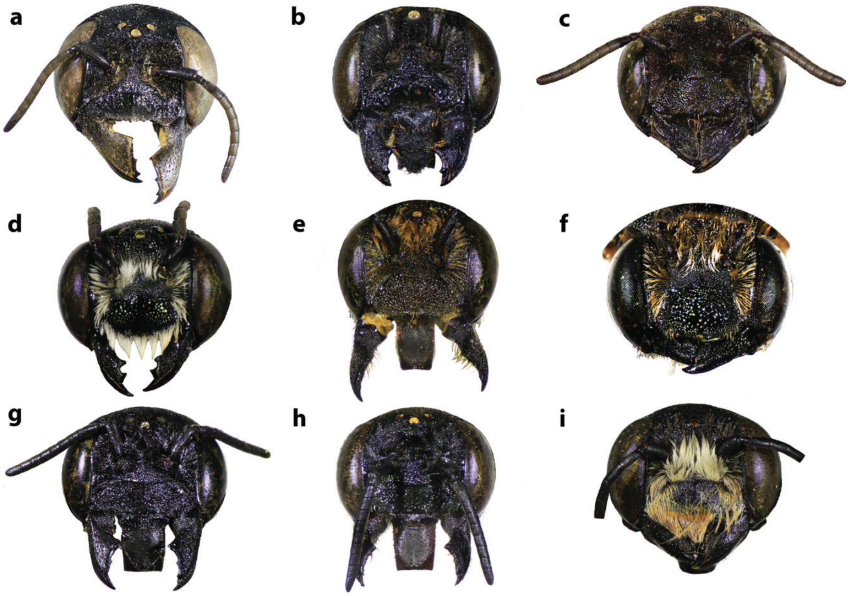


Figure 35. Frontal view of some Thai *Callomegachile* (**a–c, e, g, h** females, **d, f, i** males) **a** *M.* (*Callomegachile*) *atratiformis* **b** *M.* (*Callomegachile*) *chiangmaiensis* sp. nov. **c, d** *M.* (*Callomegachile*) *disjuncta* **e, f** *M.* (*Callomegachile*) *faceta* **g** *M.* (*Callomegachile*) *fulvipennis* **h** *M.* (*Callomegachile*) *impressa* **i** *M.* (*Callomegachile*) *memecylonae*.

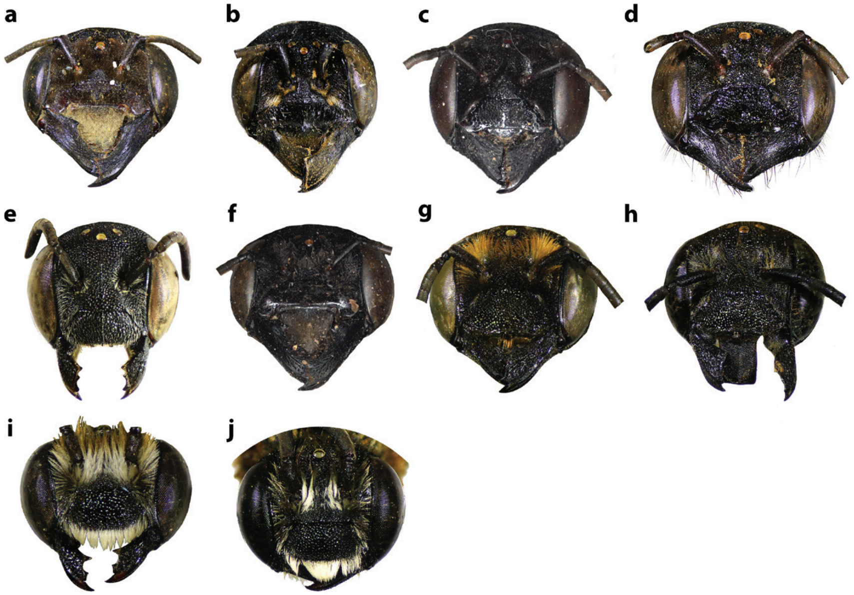


Figure 36. Frontal view of some Thai *Callomegachile* (**a–h** females, **i, j** males) **a** *M.* (*Callomegachile*) *monticola* **b** *M.* (*Callomegachile*) *odontophora* **c** *M.* (*Callomegachile*) *ornata* **d** *M.* (*Callomegachile*) *parornata* **e** *M.* (*Carinula*) *stulta* **f** *M.* (*Callomegachile*) *tuberculata* **g, i** *M.* (*Callomegachile*) *umbripennis* **h** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-4620) **j** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-3654, BSRU AA-3662 and KKIC-02).

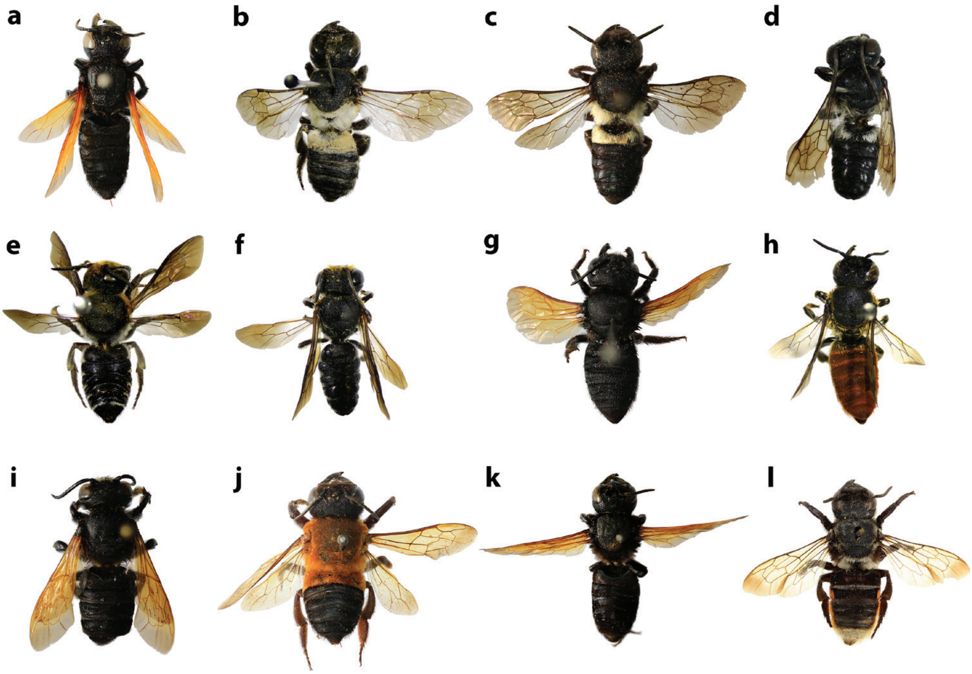


Figure 37. Dorsal view of some Thai *Callomegachile* (**a–c, e, g, h, j–l** females, **d, f, i** males) **a** *M.* (*Callomegachile*) *atratiformis* **b** *M.* (*Callomegachile*) *chiangmaiensis* sp. nov. **c, d** *M.* (*Callomegachile*) *disjuncta* **e, f** *M.* (*Callomegachile*) *faceta* **g** *M.* (*Callomegachile*) *fulvipennis* **h** *M.* (*Callomegachile*) *impressa* **i** *M.* (*Callomegachile*) *memecylonae* **j** *M.* (*Callomegachile*) *monticola* **k** *M.* (*Callomegachile*) *odontophora* **l** *M.* (*Callomegachile*) *ornata*.

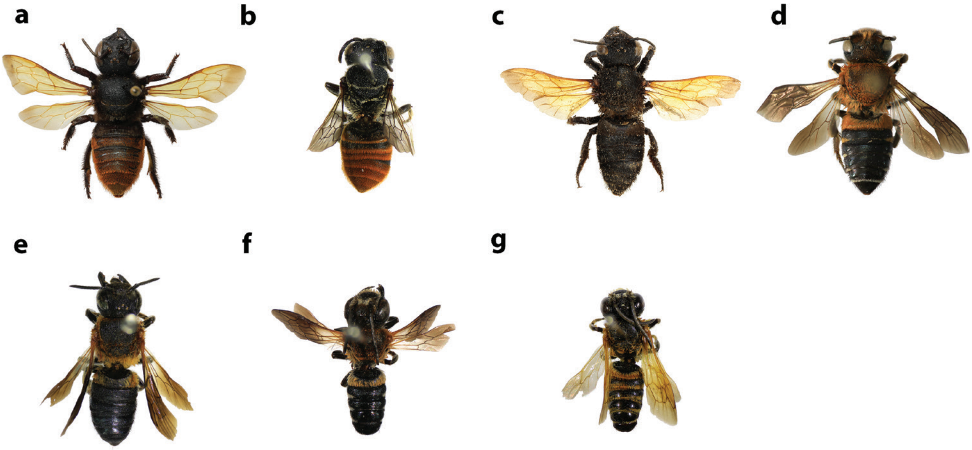


Figure 38. Dorsal view of some Thai *Callomegachile* (**a–e** females, **f, g** male) **a** *M.* (*Callomegachile*) *parornata* **b** *M.* (*Carinula*) *stulta* **c** *M.* (*Callomegachile*) *tuberculata* **d, f** *M.* (*Callomegachile*) *umbripennis* **e** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-4620) **g** *M.* (*Callomegachile*) *umbripennis* (BSRU AA-3654, BSRU AA-3662 and KKIC-02).

This is the first study to emphasize the importance of labral shape (Fig. 34a–o) for the identification in female *Callomegachile* species. In Thailand, *M.* (*Callomegachile*) *disjuncta* is the only species that has two prominent lateral teeth on distal edge of labrum (Fig. 34c), whereas in *M.* (*Callomegachile*) *fulvipennis*, *M.* (*Callomegachile*) *impressa* and *M.* (*Callomegachile*) *faceta* the lateral teeth are less prominent (Fig. 34e, f, d). *Megachile* (*Callomegachile*) *umbripennis* in Thailand has smooth and slightly concave distal edge of labrum (Fig. 34j). *Megachile* (*Callomegachile*) *parornata* has convex distal edge of labrum and *M.* (*Callomegachile*) *chiangmaiensis* sp. nov. has a distinct distal edge: medially convex with large lateral teeth (Fig. 34b). Further investigation into the applicability of the labral distal edge as diagnostic character in other *Callomegachile* species should be carried out.

Key to female species of subgenera *Callomegachile* and *Carinula* in Thailand

1 Pronotum, mesoscutum, and scutellum covered with white and/or black hairs **2**

– At least pronotum or mesoscutum and scutellum covered and/or fringed with fulvous hairs **12**

2 Most metasomal terga covered with red brick hairs or covered with black hairs with fringe of red brick hairs on lateral areas of T2–T5 (Figs 20a, 22d); scopa brick-red **3**

– Most metasomal terga covered with black hairs (sometimes T1 with band of white hairs (Figs 2a, 4a); scopa black **6**

3 Second spine of pro- and mesotibiae bifurcate (Figs 20c, 22f) **4**

– Second spine of pro- and mesotibiae not bifurcate **5**

4 T1–T4 covered with black hairs, T2 with small patch of brick-red hairs laterally; T5–T6 covered with pale light yellow hairs (Fig. 20e) ..... ***M. ornata***

– T1 covered with black hairs; T2–T5 cover with brick-red hairs; T6 covered with light yellow hairs (Fig. 22g, h) ..... ***M. parornata***

5 Clypeal margin with two small tubercles; mandible five teeth with two stout apical teeth and three small teeth basally ..... ***M. impressa***

– Clypeal margin crenulate with median carina (Fig. 33b, c); mandible four teeth ..... ***M. stulta***

6 T1 and propodeum covered with white hairs **7**

– T1 and propodeum covered with black hairs **8**

7 Clypeus with prominent apical impression and strong median carina (Fig. 31e); mandible with three stout teeth at apex; labrum oblong, apical margin medially strongly pointed with two lateral teeth (Fig. 31f) ..... ***M. chiangmaiensis* sp. nov.**

– Clypeus without apical impression, apical margin smooth with two tubercles (Fig. 4d); mandible five teeth with two stout apical teeth at apex and three small teeth basally; labrum rectangle, apical margin truncate with two lateral teeth (Fig. 4e) ..... ***M. disjuncta***

8 Base of clypeus with large protruding tubercle (Fig. 16b); mandible three teeth with small tubercle at base .... ***M. tuberculata***

– Base of clypeus without large protruding tubercle; mandible four to five teeth without small tubercle at base **9**

9 Mandible with five teeth with two stout apical teeth at apex and three small teeth basally; medium size (15–16 mm) .... ***M. fulvipennis***

– Mandible with four teeth; large size (20–23 mm) **10**

10 Apical margin of clypeus with small median tubercle (Fig. 18e); labrum oblong, apical pointed with two lateral teeth (Fig. 18d) .... ***M. odontophorum***

– Apical margin of clypeus without median tubercle; labrum rectangle, apical margin truncate without teeth **11**

11 Mesoscutum with strong transverse wrinkle pattern on disc, posteriorly with irregular punctures .... ***M. memecylonae***

– Mesoscutum with weak transverse wrinkle pattern on disc, posteriorly with weakly transverse wrinkle pattern-like disc .... ***M. atratiformis***

12 Fulvous or black tergal hair bands; base of clypeus with large protruding tubercle (Fig. 16b); mandible three teeth with small tubercles at base (Fig. 16c); scopa black; large size (20–26 mm) .... ***M. monticola***

– White tergal hair bands, sometimes interrupted at median, base of clypeus without large protruding tubercle; mandible five teeth with two stout apical teeth at apex and three small teeth basally without tubercle at base; scopa white with black at apex; median size (10–13 mm) **13**

13 Vertex with median carina (Fig. 7e); only pronotum covered with fulvous hairs; propodeal triangle with tuft white hairs .... ***M. faceta***

– Vertex without median carina; pronotum, mesoscutum, and scutellum covered with dense fulvous hairs throughout; propodeal triangle with dense fulvous hairs .... ***M. umbripennis***

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RESEARCH ARTICLE

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First discovery of the genus *Himalopenetretus* (Coleoptera, Carabidae, Patrobini) in China, with description of a new species

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Abstract

The Western Himalayan genus *Himalopenetretus* is firstly recorded from China, with one new species described, *H. burangensis* **sp. nov.** (type locality: Burang county, Xizang).

Keywords

Himalaya range, new species, Patrobini, Tibet

Introduction

Himalopenetretus Zamotajlov (Patrobini, Deltomerina) is a small genus previously containing only two species known from India and Pakistan (Löbl and Löbl 2017). This genus was erected for *H. franzi* (Zamotajlov & Sciaky, 1998) and *H. falciger* (Heinz & Ledoux, 1990), previously considered as belonging to the genus *Ledouxius*, based on the results of a phylogenetic analysis (Zamotajlov 2002). *Himalopenetretus* can be distinguished from the related genera mainly by the strongly elongated mandibles without an apical tooth.

In the summer of 2019, an expedition to Western Xizang was undertaken, during which a special specimen of Patrobini was collected in Burang County, near the border between China, Nepal and India. It was found under a rock in an alpine meadow near a glacier, together with several individuals of a small *Amara* species. The specimen was readily recognized as a new species of the genus *Himalopenetretus*. It proved to be most similar to *H. franzi*, distributed in Gangotri (North-West India), about 200 km west of the type locality of the new species. This is the first record of the genus *Himalopenetretus* from China. The main purpose of this paper is to record the genus from China and describe the new species; in addition, relationships between species of *Himalopenetretus* are briefly discussed.

Materials and methods

The holotype and only examined specimen of the new species is deposited in the collections of the Institute of Zoology, Chinese Academy of Sciences, Beijing, China (**IZAS**). The methods of dissection, illustrations, and measurements mainly follow Shi et al. (2013).

Abbreviations of measurements used in the paper are as follows: L: overall length from apex mandibles to apex of elytra, measured along elytral suture; HW: width of head, as greatest transverse distance of head; PL: length of pronotum, as linear distance from anterior to basal margin, measured along the midline; PW: width of pronotum, as greatest transverse distance of pronotum; EL: length of elytra, as linear distance from basal ridge to apex, measured along elytral suture; EW: width of elytra, as greatest transverse distance of the two closed elytra.

Taxonomy

Himalopenetretus Zamotajlov, 2002: 98

Type species. *Ledouxius franzi* Zamotajlov & Sciaky, 1998

Generic characters. The genus can be identified by the following combination of character states: a medium sized Patrobini with dorsal side reddish brown; body elongate with inconspicuous eyes; first antennomere plurisetose, with one seta distinctly longer than the others; head with four to six pairs of setae between eyes and neck constriction; mandibles strongly elongated, apical tooth absent; submentum with two pairs of setae; lateral margins of pronotum with two to five pairs of setae before middle; elytra without scutellar pore, discal setiferous pores only present on the third interval; fifth meso- and metatarsomeres glabrous or with only a few very minute setae ventrally; apical lamella of aedeagus narrow, nearly straight in lateral view and more or less twisted leftwards in dorsal view, without a tooth or other apical protuberances; endophallus with two groups of copulatory pieces: proximal one near middle, with sharp and hooked apex and a rudimentary flagellum; distal one near end of apical orifice, smaller and bilobed.

Himalopenetretus is most similar to the genus *Ledouxius*, as follows: first antennomere plurisetose; submentum with two pairs of setae; lateral margins of pronotum plurisetose; fifth meso- and metatarsomeres without long setae ventrally. However, it can be differentiated from this genus by: (1) head with four to six pairs of setae between eyes and neck constriction; (2) mandibles strongly elongated; (3) eyes less prominent, pronotum subcordate to nearly quadrate; (4) elytra without scutellar pore; (5) apical lamella of aedeagus narrow and more or less twisted leftwards in dorsal view, endophallus possessing a rudimentary flagellum on proximal copulatory pieces. The subgenus *Hasarotretus* of the genus *Ledouxius* especially resembles *Himalopenetretus* in the absence of apical teeth on the mandibles, but is different in the many important characters mentioned above.

Besides with *Ledouxius*, species of *Himalopenetretus* also share some characters with the genus *Deltomerodes*, including the extremely long temporae, the leftwards twisted apical lamella of the aedeagus, and basal part of proximal copulatory piece forming a flagellum. However, the latter genus is different in: (1) mandibles normal, not elongated; (2) head with two to four pairs of setae between eyes and neck constriction; (3) pronotum somewhat elongate and flask-shaped, lateral margins with only one (rarely two) pairs of setae before middle; (4) elytra with additional discal setiferous pores present on the fifth or seventh intervals.

Himalopenetretus includes the following three species occurring in the Western Himalayas (Map 2):

Himalopenetretus falciger (Heinz & Ledoux, 1989): Pakistan (Gilgit);
Himalopenetretus franzi (Zamotajlov & Sciaky, 1998): India (Uttar Pradesh, Tapovan);
Himalopenetretus burangensis sp. nov.: China (Xizang, Burang).

***Himalopenetretus burangensis* sp. nov.**

http://zoobank.org/1A44B1F4-43D7-4F6D-8FCF-F66249B06181

Type locality. China, Xizang: Burang County.

Type material. ***Holotype*:** male (IZAS), "Xizang, Ngari Pref., Burang County, Burang Town. N of Tinkar pass, alpine meadow. 30.2085N, 81.0661E, 4800m."; "under stone, 2019. VII. 15, Shi HL lgt., Exp. BJFU 2019"; "DNA sample series number SHL2019--Xiz006"; "HOLOTYPE♂ *Himalopenetretus burangensis* sp. nov., des. YAN & SHI, 2020" [red label].

Diagnosis. The new species is diagnosable in the genus by: tarsomeres dorsally glabrous; pronotum subquadrate, lateral margins straight before posterior angles, which are a little pointed, disc with a pair of small concavities to the side of the median line, lateral margins with two or three pairs of setae before middle; apical lamella of aedeagus elongate and evidently twisted leftwards, apex a little capitate and hooked leftwards.

The new species is most similar to *H. franzi*, but the latter species is different from it in: pronotum subcordate, lateral margins evidently sinuate before posterior angles, disc without concavities, lateral margins with four or five pairs of setae before middle; apical lamella of aedeagus shorter, only very weakly twisted leftwards, apex simple, narrowly triangular. The other species of the genus, *H. falciger*, is quite different from these two species in having tarsi dorsally pubescent and in many other respects.

Description. Habitus as in Figure 1. Medium-sized for a Patrobini species (L = 9.2 mm; EW = 2.9 mm).

General appearance: Dorsal side reddish brown, shiny, without metallic luster; head, mandibles and pronotum brown, elytra a little lighter; antennae, palpi and legs light reddish-brown; ventral side largely dark brown, abdominal sternum reddish-brown. Dorsal side glabrous and smooth except for lateral margins and basal foveae of pronotum punctate. Microsculpture invisible on head and pronotum, isodiametric on elytra, hardly visible near base, strong in apical third.

Head: Broad and ovate; surface smooth, without prominent punctures. Mandibles strongly elongated, narrow, without an apical tooth (Figs 2, 3); apical maxillary palpomere broadest in middle, penultimate and antepenultimate ones with ring of apical setae; ligula with two apical setae. Antennae pubescent from third antennomere; first antennomere plurisetose, with one seta distinctly longer than the others; second antennomere glabrous except subapical ring of setae (Fig. 5). Eyes very small, inconspicuous, not convex; temporae long, much longer than eye diameter, moderately tumid; neck constriction rather shallow. Frontal furrows short but very deep, extended to level of posterior edge of eyes, slightly divergent posteriorly, almost parallel; with five or six setae between eyes and neck constriction on each side (holotype with six on the left side and five on the right side), including: one supraorbital seta near level of middle of eyes, three setae between posterior margins of eyes and neck constriction, one or two setae adjoining to neck constriction. Mentum basally with two rather deep longitudinal foveae, tooth narrow and bifid, apical notch shallow; submentum with two setae on each side (Fig. 6).

Pronotum: Nearly quadrate, a little broad, PW/PL = 1.24, faintly wider than head, PW/HW = 1.23, widest near anterior third, fairly convex, moderately constricted posteriorly. Anterior margin nearly straight; lateral margins fairly rounded in front, without prominent sinuation before posterior angles; posterior margin nearly straight; anterior angles rounded but distinct, slightly protruding anteriorly; posterior angles near rectangular, apex slightly pointed outwards. Anterior transverse impression shallow, sparsely punctate; basal foveae shallow, coarsely punctate and wrinkled; disk smooth, with a pair of shallow concavities to the side of median line (Fig. 4); median line distinct, almost reaching both extremities; lateral grooves coarsely punctate. Lateral margins each with two or three setae before middle (holotype with two setae on the left and three setae on the right); one pair of setae situated a little before posterior angles.

Elytra: Oblong-ovate, depressed; EL/EW = 1.66, widest near posterior third; humeri narrowly rounded, humeral tooth indistinct; lateral margins fairly widened and flat. Intervals slightly convex; striae well incised, finely punctate basally; scutellar stria

Xizang, Ngari Pref., Burang County, Burang Town, N of Tinkar Pass, alpine meadow N30.2085 E81.0661, 4800m

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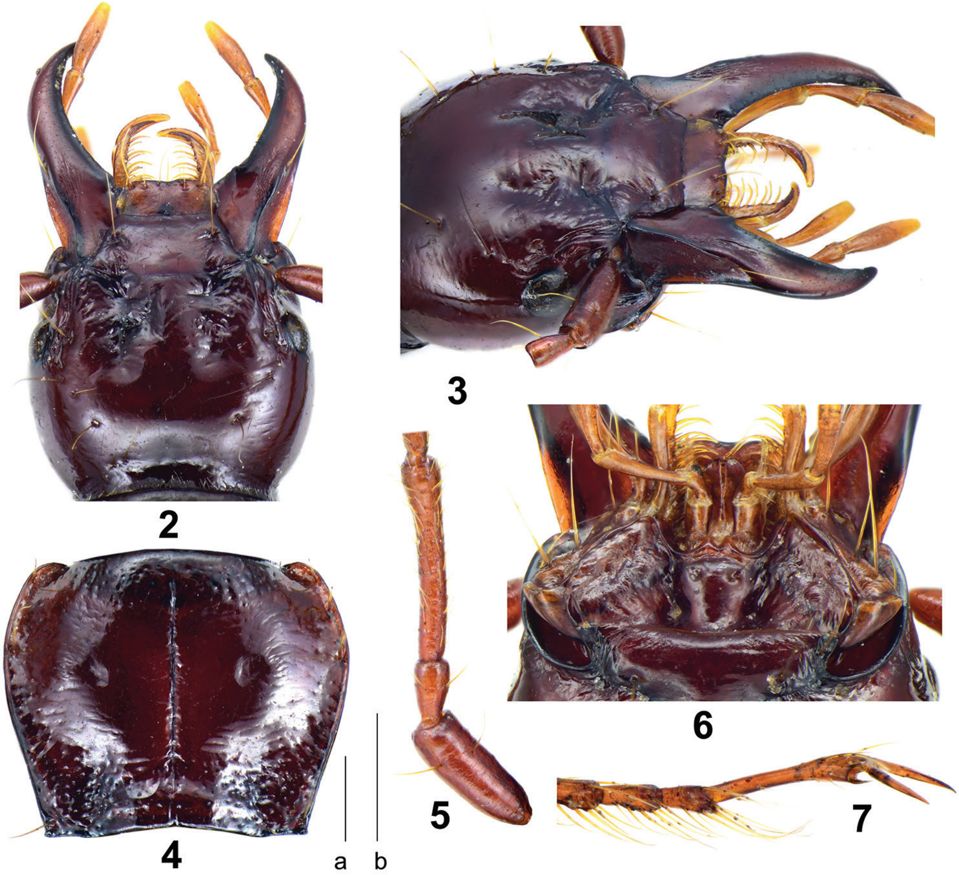
DNA sample
series number
SHL2019--**Xiz006**

HOLOTYPE ♂
Himalopenetretus burangensis **sp.nov.**
des. Yan & Shi, 2020

Figure 1. *Himalopenetretus burangensis* sp. nov. (holotype male, IZAS). Habitus and labels. Scale bar: 1.0 mm.



Figures 2–7. *Himalopenetretus burangensis* sp. nov. (holotype male, IZAS). **2** Dorsal side of head **3** Lateral side of head **4** Dorsal side of pronotum **5** First three segments of antennae **6** Labium **7** Lateral side of metatarsomeres. Scale bars: 0.5 mm (**a** for **2–4**, **b** for **5–7**).

long, situated between elytra suture and first striae, apex free, scutellar pore absent; third interval with five setiferous pores, all adjoining third stria, the first one near basal fifth; fifth interval with one setiferous pore near apex; umbilicate series on ninth interval composed of eight to ten pores, nearly equally arranged, a little denser in posterior areas.

Ventral side: Prosternum smooth, propleuron densely punctate; mesosternum and mesopleuron wrinkled, mesopleuron with sporadic coarse punctures; mesepimeron narrow, slightly widened laterally, suture separating mesepisternum and mesosternum joining lateral margin of metasternum; metepisternum rather long and narrow, not punctate. Lateral areas of abdominal sternites slightly rugose, abdominal sternite IV to VI with two setae near middle on each side; VII with one or two setae on each side in male (holotype with two setae on the left and one seta on the right).

Legs: Males with the first two protarsomeres slightly expanded, the second protarsomere distinctly wider than the third one which nearly triangular; the fourth protar-

Figures 8–13. *Himalopenetretus burangensis* sp. nov. (holotype male, IZAS). **8** Right paramere of aedeagus **9** Left paramere of aedeagus **10–13** Median lobe of aedeagus, right lateral view (**10, 12**), ventral view (**11, 13**) **12, 13** were captured after the genitalia were treated with a 10% KOH solution for 12h to show features of the endophallus. Scale bar: 0.5 mm.

somere evidently bilobed; metatrochanter normal, not protruding or exceeding lateral margin of body; tarsomeres generally glabrous dorsally, only with a few very minute setae; the fifth meso- and metatarsomeres generally glabrous ventrally, with one to three pairs of minute setae (Fig. 7).

Male genitalia (Figs 8–13): Median lobe of aedeagus strongly bent at base, gutter-shaped and opened dorsally; in lateral view apical lamella nearly straight; in dorsal view

Map 1. Distribution map for *Himalopenetretus burangensis* sp. nov. (red).

apical lamella very elongate and narrow, gradually attenuated towards apex and prominently twisted leftwards, apex capitate, forming a faint hook to the left. Armature of endophallus consisting of two groups of copulatory pieces: proximal one near middle with sharp and hooked apex and a rudimentary flagellum; distal one near end of apical orifice, smaller and bilobed. Left paramere larger than right one, both short but sharply contracted towards apex, each with two long apical setae.

Female unknown.

Distribution. This species is known only from the holotype from Burang, Xizang (Map 1).

Etymology. The new species is named after its type locality, Burang County.

Remarks. Among the three species of the genus *Himalopenetretus*, *H. burangensis* sp. nov. could be closest to *H. franzi* for their near distribution ranges and morphological similarities. In particular, the dorsally glabrous tarsomeres of these two species could be of taxonomic significance.

The results of a phylogenetic analysis by Zamotajlov (2002) indicate a close relationship between *Himalopenetretus* and *Ledouxius*, and the subgenus *Ledouxius* (*Hasarotretus*) has some intermediate character states between these two genera (Zamotajlov and Sciaky 2006). In general, *H. burangensis* sp. nov. fits most of the generic characters of *Himalopenetretus* previously suggested (Zamotajlov 2002), but the lateral margins of pronotum with two or three setae before middle make the new species somewhat resemble the genus *Ledouxius*. However, from its general habitus, male genitalia, and many other respects, the new species clearly belongs to the genus *Himalopenetretus*.

Map 2. Distribution map for *Himalopenetretus* species: *H. burangensis* sp. nov. (red); *H. franzi* (blue); *H. falciger* (magenta).

Thus, the generic diagnosis of this genus should be slightly modified to include the new species, as follows: pronotum lateral margins with two to five setae before middle.

The discovery of this new species extends the eastern limit of the known distribution of the genus *Himalopenetretus*. Considering that all the species of this genus are very rare, and that explorations in high mountains of the Western Himalayas are relatively inadequate, the discovery of additional new species is expected from the nearby areas, such as southwestern Xizang and Nepal (Map 2).

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