ZooKeys 254: 1–22 (2012)
doi: 10.3897/zookeys.254.3892
www.zookeys.org

RESEARCH ARTICLE

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Acoustic analysis reveals a new cryptic bush–cricket in the Carpathian Mountains (Orthoptera, Phaneropteridae)

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Academic editor: *A. Gorochov* | Received 28 November 2012 | Accepted 10 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:71E92801-035C-47AB-99E6-FAB2CFFB1B8B

Citation: Iorgu IŞ (2012) Acoustic analysis reveals a new cryptic bush–cricket in the Carpathian Mountains (Orthoptera, Phaneropteridae). ZooKeys 254: 1–22. doi: 10.3897/zookeys.254.3892

Abstract

A new morphologically cryptic species of phaneropterid bush–cricket from the genus *Isophya* is described from the Eastern Carpathian Mountains: *Isophya dochia* **sp. n.** Sound analysis and morphological details are discussed in the paper, comparing the new species with several *Isophya* species having similar morphology and acoustic behavior.

Keywords

Isophya, bioacoustics, taxonomy, Carpathians

Introduction

One of the largest and most enigmatic phaneropterid genera, with 90 species known and 45 species present in Europe, *Isophya* Brunner von Wattenwyl inhabits Southern and Eastern Europe, Asia Minor and Caucasus up to Kazakhstan, Iran and Iraq (Eades et al. 2012).

Dissimilar to morphological homogeneity in genus *Isophya*, the specific structure of male acoustic signals shows clear differences in close related species and is used as the most effective tool for identifying and clarifying taxonomic relations (Heller et al.

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2004, Chobanov 2009, Orci et al. 2010a, 2010b, Szövényi et al. 2012). Contrary to this general opinion, a recent study on several related *Isophya* species from Asia Minor suggests that, in some cases, evolutionary changes in song appear slower than in morphology (Sevgili et al. 2012). In this genus, the male–female pair is usually formed during an acoustic duet; females locate the males by phonotaxis and respond to their songs with simple impulses (Orci et al. 2001, Orci 2007, Orci et al. 2010a, 2010b) or complicated click–series (Iorgu 2012) within a species–specific time span.

The *Isophya* species are very interesting subjects to study: apart from their remarkable behavior, most of the species have restricted distribution areas and present a large number of endemics. For the time being, 16 *Isophya* species are known to occur in the Carpathian Mountains (Nagy 2005, Heller 2012). Recently, three new species have been described from the Eastern Carpathian Mountains: *Isophya sicula* Orci, Szövényi and Nagy, 2010; *Isophya ciucasi* Iorgu and Iorgu, 2010 and *Isophya nagyi* Szövényi, Puskás and Orci, 2012.

In the summer of 2005, several bush–crickets were collected from the area called "Poliţa cu Ariniş", close to the subalpine meadows of Ceahlău Mountains. Studying only the morphological characters, they were identified back then as *Isophya camptoxypha* (Fieber) and up to the summer of 2010 no acoustic data of this geographically isolated population were available. With the first recorded songs, its status had to be changed in a new taxa, perfectly morphologically cryptic: *Isophya dochia* sp. n.

Material and methods

Audio recordings were taken with an Edirol R–09HR digital recorder (microphone frequency response 20–40000 Hz, sampling rate of 96000 Hz, 24 bit amplitude resolution). In the field, we used an Edirol CS-15R unidirectional external microphone attached to the digital recorder (frequency response 200–17000 Hz). Temporal and spectral sound analyses were performed with the software Audacity 2.0.2.

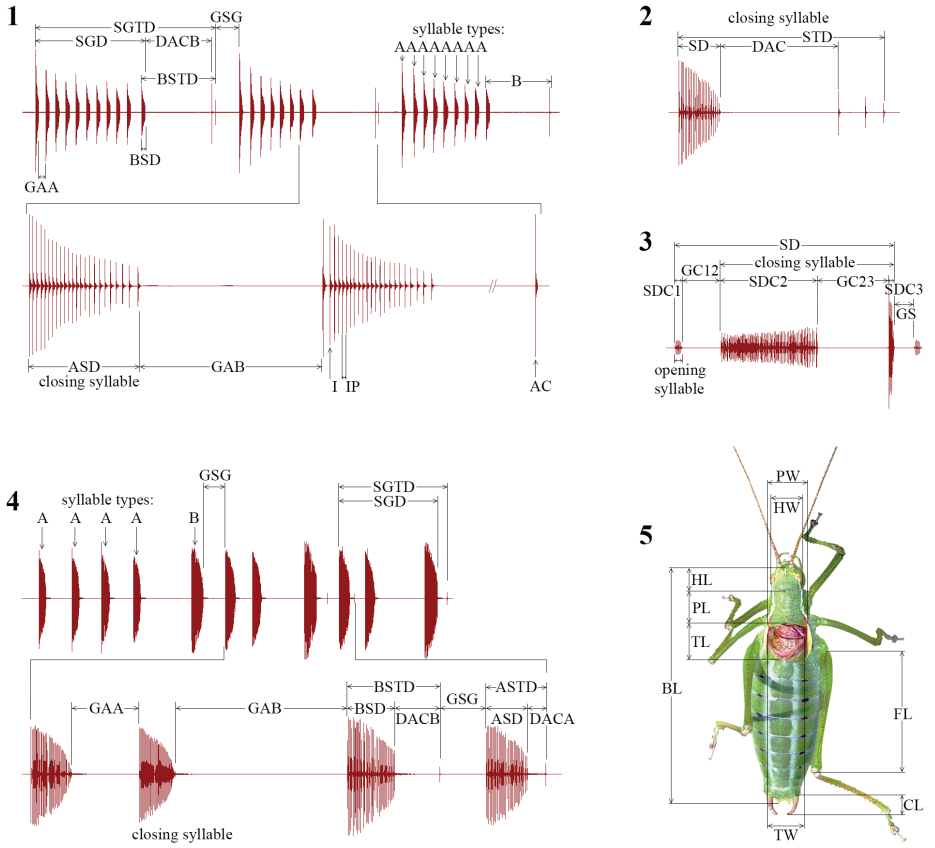
Song terminology and abbreviations are adapted from Heller et al. 2004, Orci et al. 2005 and Orci et al. 2010a (Figs 1–4; see Appendix 1: Isophya song abbreviations).

Morphological traits were examined with a stereomicroscope and the following characters were measured for 20 males and 20 females of the new species: body length (BL), head width (HW), head length (HL), pronotum maximum width (PW), pronotum length (PL), left tegmen maximum width (TW), tegmina length (TL), cercus length (CL) and femur length (FL) (Fig. 5).

Photos were taken with Canon EOS 600D DSLR camera and Canon 100 mm 1:1 and Canon MP–E 65 mm 5:1 macro lenses, using photo stacking method for morphological characters. Movements of tegmina during sound production have been video recorded with the same camera, with the external microphone attached.

The distribution area map was drawn using the altitude layer from Jarvis et al. (2008).

Type specimens are preserved in the collections of "Grigore Antipa" National Museum of Natural History, Bucharest, Romania.



Figures 1–5. Schematics of studied song (**1** *Isophya dochia* sp. n. **2** *Isophya camptoxypha* **3** *Isophya nagyi* **4** *Isophya harzi*) and morphological (**5** *Isophya dochia* sp. n.) characters.

Data resources

The data underpinning the analyses reported in this paper are deposited in the Dryad Data Repository at doi: 10.5061/dryad.256qh

Taxonomy

***Isophya dochia* sp. n.**
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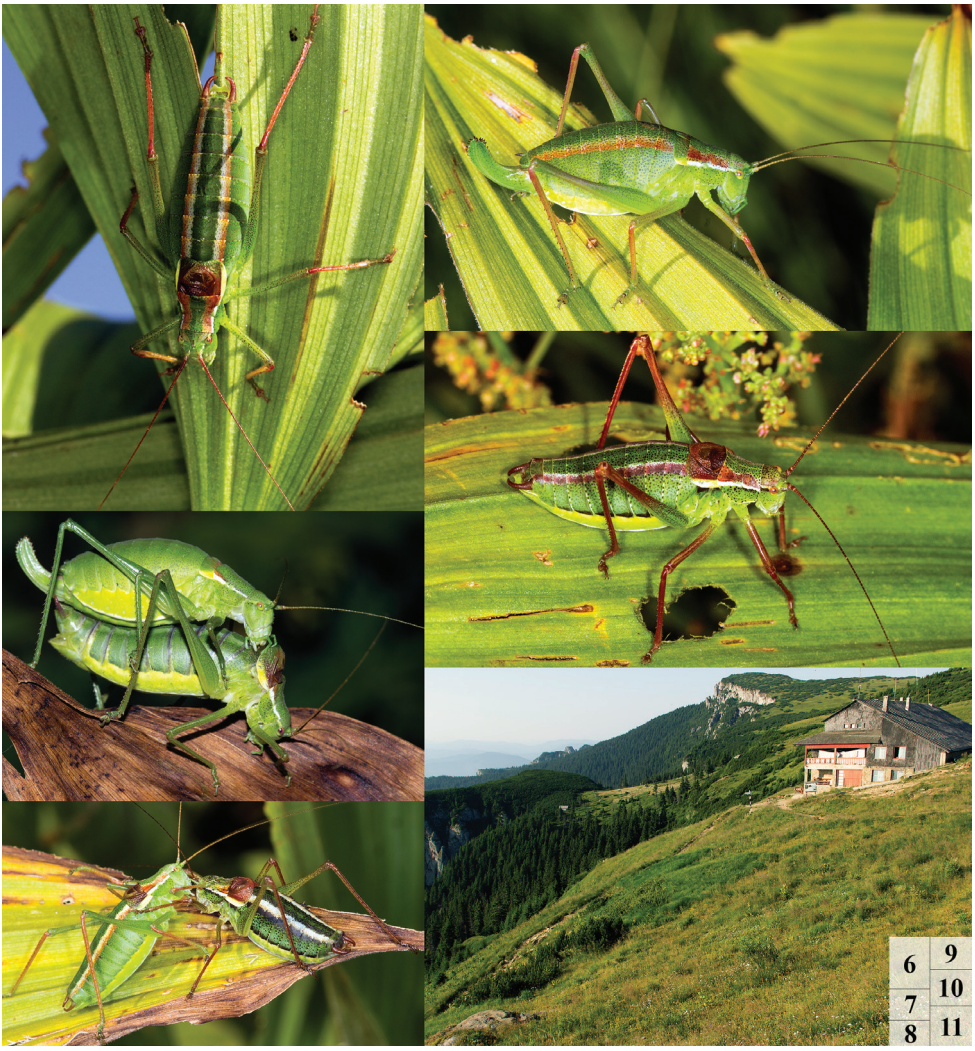
http://species-id.net/wiki/Isophya_dochia
Figs 1, 6–26, 32, 38, 44, 45, 51, 57, 63, 69, 75

Type locality. Romania, Eastern Carpathian Mountains, Ceahlău Mountains.

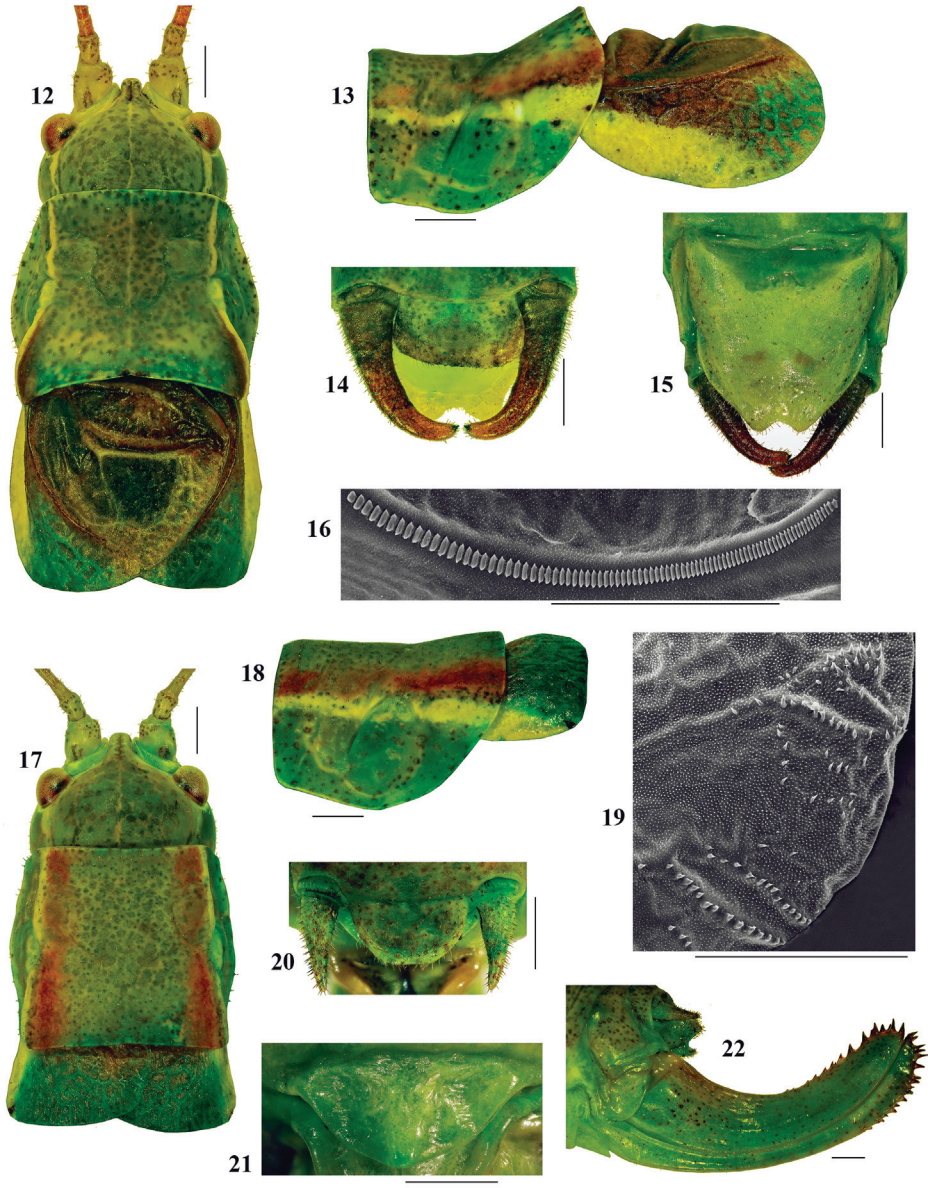
Type material. Holotype: male. Original label: "România, Munții Ceahlău, Poliţa cu Arinişi, 46°57.90'N, 25°56.32'E, alt. 1620 m, 09.08.2005, Leg. Iorgu I. Ş."



Figures 6–11. *Isophya dochia* sp. n.: **6** male habitus **7** copula **8** males' rivalry **9** female habitus **10** male habitus **11** habitat in Ceahlău Mountains, near Dochia cabin (1740 m).

Paratypes: 10 ♂♂ 7 ♀♀, labeled: "România, Munții Ceahlău, Cabana Dochia, 46°57.94'N, 25°57.02'E, alt. 1740 m, 02.07.2010, Leg. Iorgu I. Ş."; 6 ♂♂ 5 ♀♀, labeled: "România, Munții Ceahlău, Piatra Lată, 46°57.73'N, 25°56.93'E, alt. 1720 m, 10.07.2012, Leg. Iorgu I. Ş", a microSD card containing audio and video records of male song attached; coll. "Grigore Antipa" National Museum of Natural History, Bucharest, Romania.

Audio recordings. 8 ♂♂, 5 July 2010, in laboratory, air temperature 25°C; 17 ♂♂, 10 July 2012, in the field, air temperature 20°C; 5 ♂♂ 5 ♀♀, 12 July 2012, in laboratory, air temperature 24°C (see Appendix 2: Isophya dochia sp n audio and Appendix 3: Isophya dochia sp n video).



Figures 12–22. *Isophya dochia* sp. n.: **12** dorsal view of male head, pronotum and tegmina **13** lateral view of male pronotum and tegmina **14** male cerci **15** male subgenital plate **16** male stridulatory file (SEM photo) **17** dorsal view of female head, pronotum and tegmina **18** lateral view of female pronotum and tegmina **19** female stridulatory bristles (SEM photo) **20** female cerci **21** female subgenital plate **22** ovipositor. Scale 1 mm.

Comparative material (see Appendix: 4 Isophya comparative material).

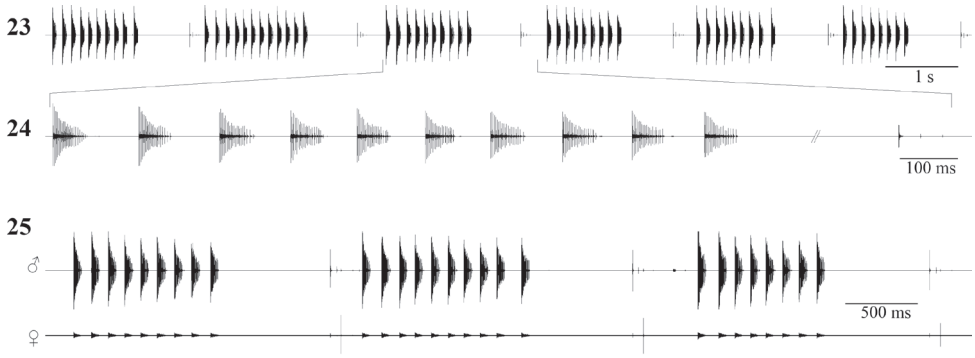
Description (Figs 12–22; Table 1). Male. Fastigium verticis slightly tapering frontward, half as wide as scapus, with a dorsal groove (Fig. 12). Head length 1.7 times the

Table 1. Studied morphological characters (see Material and Methods) O – ovipositor.

		HL (mm)	HW (mm)	PL (mm)	PW (mm)	TL (mm)	TW (mm)	BL (mm)	FL (mm)	CL (mm)	O (mm)
male	Minimum	1.87	3.19	3.34	3.89	3.8	3.83	21.1	14.57	2.4	–
	Maximum	1.91	3.45	4.02	4.25	4.43	4.29	24.6	16.38	2.6	–
	Mean	1.89	3.32	3.8	4.07	4.23	4.07	22.32	15.2	2.51	–
	S. D.	0.02	0.1	0.24	0.14	0.23	0.16	1.12	0.61	0.08	–
female	Minimum	2.79	3.64	4.02	4.14	1.68	–	22.67	14.71	1.25	8.32
	Maximum	3.07	3.98	4.58	4.66	2.39	–	26.23	16.56	1.64	8.96
	Mean	2.9	3.8	4.31	4.37	1.92	–	24.44	15.36	1.42	8.61
	S. D.	0.11	0.11	0.18	0.21	0.25	–	1.31	0.66	0.15	0.21

pronotum length and head width about 1.3 times the maximum pronotum width. Pronotum saddle shaped from a lateral view, paranota with concave dorsal margins, anterior and ventral borders straight, posterior edge moderately convex. Pronotal disc slightly constricted in the transverse sulcus area, with lateral carinae marginally divergent in mesozona and convex in metazona (Figs 12, 13). Wings as long as or slightly longer than pronotum, with reticulate venation, usually surpass the posterior edge of first abdominal tergite. Cu2 vein length about 3/4 the posterior margin of pronotum; angle between cubital veins about 70°. Speculum large and rectangular. Edge of tegmen at distal end of Cu2 vein forms an obtuse angle of about 130° (Fig. 12). During the quiet wing openings in song production, the white anterior border of tegmen is well visible, but its role has not been established yet (see Appendix 3: Isophya dochia sp n video). Stridulatory file arcuate, 2.2–2.4 mm long, counts 82–89 teeth; distal teeth larger and rarer than proximal ones (Fig. 16). Epiproct about twice as wide as long; cercus slender, narrowing towards tip, slightly curved in its apical fourth, with fine, small hairs; terminal denticle located in middle of cercus apex (Fig. 14). Subgenital plate elongated, narrowed apically, with triangular apical incision (Fig. 15). Hind femur about 4–4.3 times the pronotum length, without ventral spines. Coloration green, densely punctuated with fine, dark green and brown spots. Several males with two dorso–lateral, parallel stripes from pronotum to end of abdomen, red, orange, white, violet or yellow colored. Antennae greenish–brown or reddish–brown, with light brown or green scapus. Compound eyes bicolor: upper part brownish–red and lower part green. A yellowish or white band begins behind the eye and ends at posterolateral angle of wing. Tegmina brown, dark brown or dark red, apically green and costal margin greenish–white or yellowish–white. Cerci brown or reddish–brown, green at base. Ventral side of body yellowish–green. Femora, tibiae and tarsi usually green, brownish or reddish.

Female. Fastigium roughly as in male (Fig. 17). Head length 1.7 times the pronotum length and head width about 1.3 times the maximum pronotum width. Pronotum disc marginally enlarged in its posterior part, with straight lateral carinae, paranota as in males (Figs 17, 18). Wing with dense reticulate venation, surpass the posterior margin of first abdominal tergite. Stridulatory bristles located on cubital veins in the inner latero–posterior part of right tegmen (Fig. 19). Cercus short, hairy, conical (Fig.



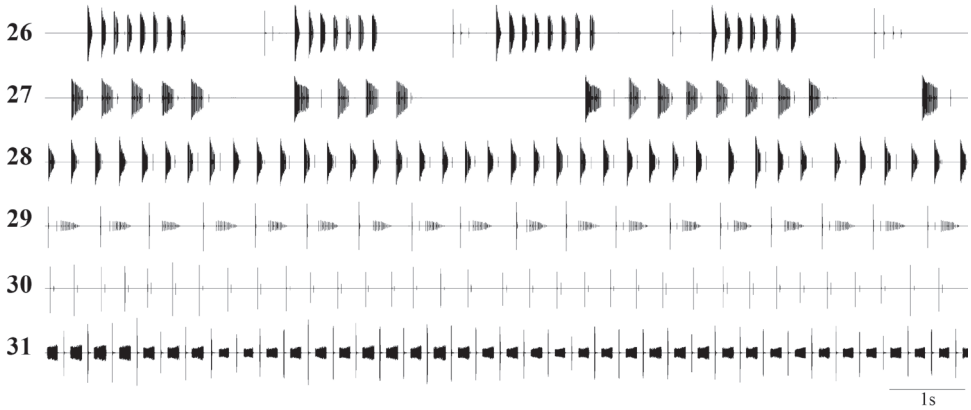
Figures 23–25. Oscillographic sound analysis in *Isophya dochia* sp. n., Ceahlău Mountains (24°C): **23** male song, consisting of syllable groups **24** detailed group of syllable **25** male–female mating acoustic duet.

20). Subgenital plate rounded, narrow, about twice as wide as long (Fig. 21). Ovipositor short, upcurved, 1.9–2.1 times the pronotum length, upper margin with 9–10 denticles and lower margin with 8–9 denticles (Fig. 22). Hind femur 3.5–3.7 times the pronotum length, without ventral spines. Body coloration as in males, wings light brown or green–yellowish, ovipositor green.

Bioacoustics. Males stridulate at dusk and during the night, rarely during daytime. The tegmino–tegminal stridulation consists of groups of 5–18 syllables (mean±SD: 8.9±3.48, n=30 ♂♂). A group lasts for 1288–4761 ms and successive groups are separated by an interval of 157–326 ms. Groups have a repetition rate of about 10–25 per minute, depending on the number of syllables.

Two types of syllables may be observed in a group: "A" type and "B" type. Both syllable types are produced when the male closes its tegmina. The song pattern may be formulated as "A...AB–A...AB–A...AB–A...AB" and so on, where the unit "A...AB" forms a distinct group of syllables, "..." means a variable number of "A" syllables and "–" means the interval between successive groups of syllables. The "A" type syllable is formed of a compact series of 9–29 impulses (mean±SD: 18.6±6.31), lasting for 30–70 ms (mean±SD: 50.74±12.83). The "B" type syllable is formed of a compact series of 9–15 impulses (mean±SD: 12.36±2.1), lasting for 30–59 ms (mean±SD: 39.3±9.4) and always followed by a series of 1–4 after–clicks at an interval of 621–1655 ms (mean±SD: 1109.1±212.65). The production of the last syllable ("B" type) in each sequence is complex: the male partially closes tegmina, then holds them half–closed for about 621–1655 ms, and finally completely closes the wings with the after–clicks. This late production of after–clicks may function as the trigger element for female acoustic answer. Another possibility is that longer silent gaps may help the male to save up energy or simply listen to its environment, in order to detect possible threats or other singing males (Orci et al. 2010b, Szövényi et al. 2012).

The impulse interval is about 3–6 ms in all syllables and the acoustic signal slowly decreases in amplitude from beginning to end. In a group, the following syllable be-



Figures 26–31. Oscillographic sound analysis: **26** *Isophya dochia* sp. n., Ceahlău Mountains (24°C) **27** *Isophya harzi*, Cozia Mountains (25°C) **28** *Isophya camptoxypha*, Vânători Neamț (26°C) **29** *Isophya ciucasi*, Ciucaş Mountains (26°C) **30** *Isophya sicula*, Harghita Mountains (25°C) **31** *Isophya nagyi*, Călimani Mountains (25°C).

gins 60–131 ms (mean±SD: 96.28±24.62) later (Figs 23, 24). The carrier wave has the strongest components between 20–40 kHz, with the highest peak at about 29 kHz.

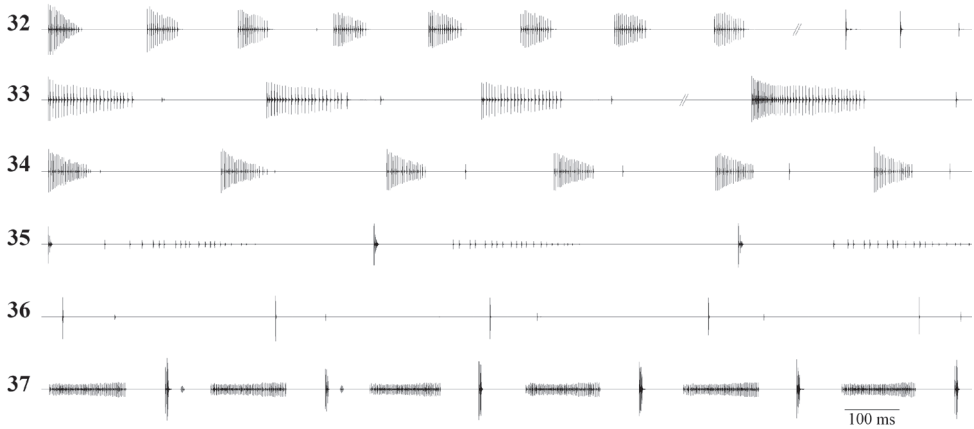
Females find males by phonotaxis and if willing to mate with the singing male (Fig. 7), they produce isolated impulses. In the resulting male–female duet, the female stridulates only after the male after–clicks (ending part of "B" syllable), with a latency of 13–34 ms (mean±SD: 25.66±6.42, n=12 responses from 3 females) (Fig. 25).

Distribution and ecology. *Isophya dochia* sp. n. populates mesophytic subalpine meadows at about 1600–1900 m, in Ceahlău Mountain Massif, Eastern Carpathians (Fig. 11). The specimens were collected from leaves of *Urtica*, *Rubus*, *Veratrum, Rumex, Aconitum, Vaccinium, Hypericum, Stachys, Junniperus* etc. Few other bush–crickets and grasshoppers were found occurring simpatrically with the new species: *Metrioptera bicolor* (Philippi), *Metrioptera brachyptera* (Linnaeus), *Pholidoptera transsylvanica* (Fischer), *Miramella ebneri* (Galvagni), *Euthystira brachyptera* (Ocskay), *Myrmeleotettix maculatus* (Thunberg), *Chorthippus biguttulus* (Linnaeus), *Chorthippus parallelus* (Zetterstedt) etc. The bush–cricket *Isophya dochia* sp. n. has the same phenology as other subalpine *Isophya* species: female lays her eggs isolated in holes bitten in broad leaves of *Urtica*, *Rubus* etc. Eggs pass the winter in the litter and larvae hatch in late spring, after the snow melts in the high mountains. Depending on weather, first adults exuviate in late June and live up to August.

Etymology. A noun in apposition; from the name of Dochia, a Romanian legendary female character based on an earlier deity of land and agriculture from the Dacian pantheon, and that of the eponymous rock in Ceahlău Mountains.

Discussions (see Appendix 5: *Isophya harzi*, Appendix 6: *Isophya camptoxypha*, Appendix 7: *Isophya ciucasi*, Appendix 8: *Isophya sicula*, Appendix 9: *Isophya nagyi*).

Discovery of the bush–cricket *Isophya dochia* sp. n. is surprising, especially as it is a morphological cryptic species closely related to *I. camptoxypha*. Some other recently

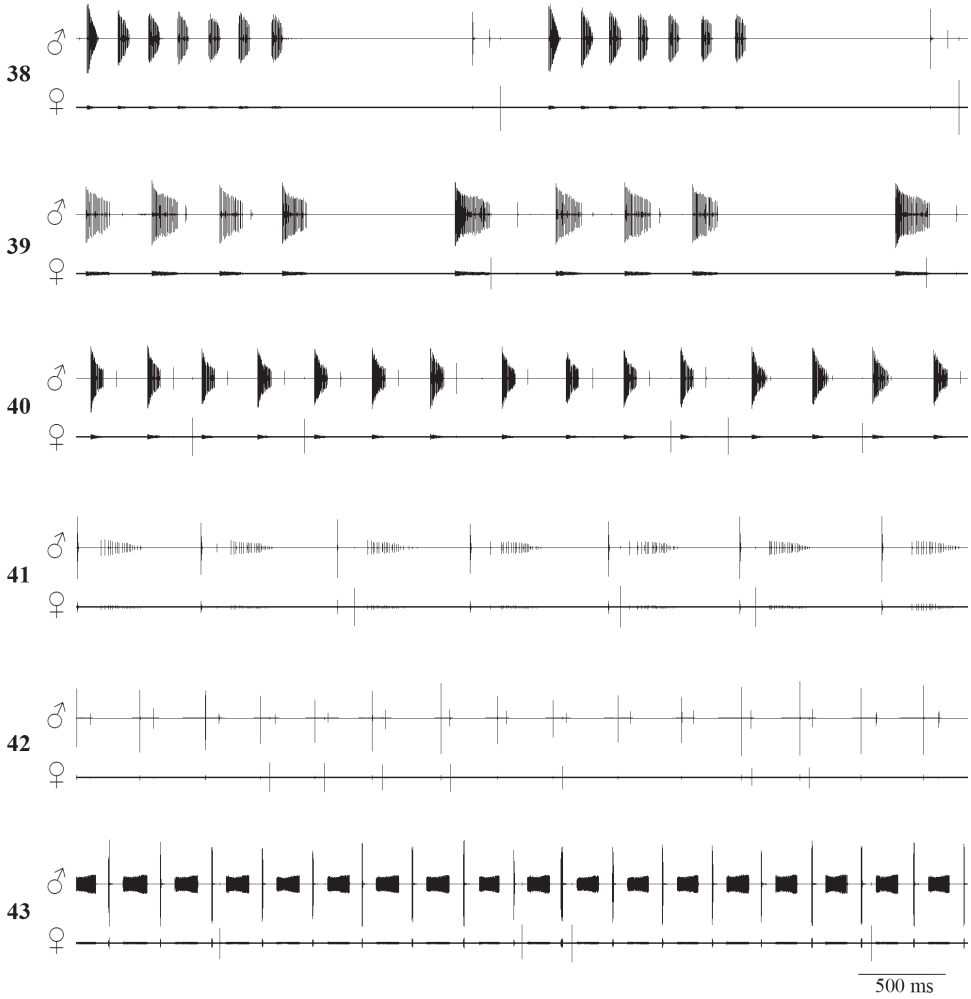


Figures 32–37. Detailed syllables: **32** *Isophya dochia* sp. n. **33** *Isophya harzi* **34** *Isophya camptoxypha* **35** *Isophya ciucasi* **36** *Isophya sicula* **37** *Isophya nagyi*.

described species, *Isophya ciucasi*, *Isophya nagyi* and *Isophya sicula*, show high resemblance in morphology, while *Isophya harzi* Kis has a similar calling song (Figs 26–37). The song of these six species can be readily distinguished as syllables grouped in short sequences (*Isophya dochia* sp. n. and *Isophya harzi*) or arranged in series (*Isophya camptoxypha*, *Isophya ciucasi*, *Isophya nagyi* and *Isophya sicula*).

Both *Isophya dochia* sp. n. and *Isophya harzi* stridulate well defined assemblies of syllables (Figs 26, 27, 32, 33). Two other species have similar song patterns: *Isophya posthumoidalis* Bazyluk, distributed in Poland, Slovakia and N Romania (Szövényi and Orci 2008) and *I. beybienkoi* Mařan, known so far only from a very small area in SE Slovakia. In all these four species, males produce long sequences in which two types of syllables may be noticed: "A" and "B", the rhythm of the whole song being a constant repetition that may be formulated as "A...AB–A...AB–A...AB" (in *Isophya dochia* sp. n. and *I. harzi*), "A...A–B–A...A–B–A...A" (in *I. posthumoidalis*) and "A...A–BA...A–BA...A" (in *I. beybienkoi*). The descriptions of calling songs of *Isophya posthumoidalis* and *I. beybienkoi* (Orci et al. 2001, Heller et al. 2004, Orci et al. 2010a) suggest that syllable types are very different from *I. dochia* sp. n.: in *I. posthumoidalis*, syllable "A" consists of a compact series of 5–10 impulses, lasting for 12–15 ms, and syllable "B" is a single impulse followed by 1–3 after–clicks, while in *I. beybienkoi* syllable "A" is formed of 5–13 impulses, lasting for 10-32 ms, and syllable "B" consists of a compact series of 2–9 short impulses followed by a longer one and 1–3 after–clicks. Not possessing any personal data on these two species, their songs and morphology were not illustrated in present paper.

In *Isophya camptoxypha*, the syllable is a short and compact series of impulses, lacking or followed by one (extremely rare 2–7) after–click (Figs 28, 34). Males of *Isophya ciucasi* stridulate a shorter syllable, but followed by a very high number of after–clicks, usually 10–30 (Figs 29, 35). *Isophya sicula* produces the shortest known syllables within this genus, consisting of only 1–3 impulses, missing or followed by one (extremely rare 2–5) after–click (Figs 30, 36). Finally, the song of *Isophya nagyi* is the most in-



Figures 38–43. Male–female duet: **38** *Isophya dochia* sp. n., Ceahlău Mountains (24°C) **39** *Isophya harzi*, Cozia Mountains (25°C) **40** *Isophya camptoxypha*, Pleşa (24°C) **41** *Isophya ciucasi*, Ciucaş Mountains (24°C) **42** *Isophya sicula*, Harghita Mountains (25°C) **43** *Isophya nagyi*, Călimani Mountains (25°C).

teresting, syllables being divided in two or three distinct fragments: the first one is an opening syllable and last two are part of the same closing syllable (Figs 31, 37). During our studies, less than 1% of analyzed syllables contained all three components, males usually producing sounds only when closing the tegmina.

Another interesting feature of the song in some of these species is the similarity of time–windows when after–clicks are produced: usually 50–80 ms after the syllable, but up to 151 ms in *I. camptoxypha* (Heller et al. 2004) and up to 110 ms in *I. sicula*. After–clicks may also follow both types of syllables of *Isophya harzi*. However, we observed a longer after–click delay (75–202 ms) next to the "B" syllable of the sequence

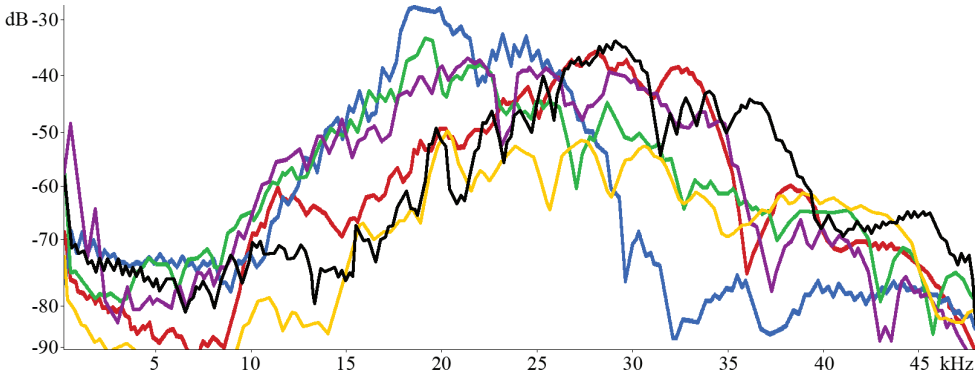


Figure 44. Sound spectrum: black – *Isophya dochia* sp. n.; blue – *Isophya harzi*; red – *Isophya camptoxypha*; green – *Isophya ciucasi*; yellow – *Isophya sicula*; purple – *Isophya nagyi*.

and a shorter delay after the "A" syllables (40–90 ms). The longest after–click delay is found in *I. dochia* sp. n., up to 1655 ms.

A remarkable interspecific variation in this group is the number of teeth on male stridulatory file (Figs 63–68), which may be correlated with the total length and number of impulses in the unit syllable + gap + after–clicks, meaning a total closing stroke of wing. In the species that sing well defined groups of syllables: in *Isophya harzi*, 98–130 pegs produce a total number of 37–59 impulses lasting for 94–418 ms in the "B" syllable and 23–55 impulses lasting for 75–258 ms in the "A" syllable, while in *Isophya dochia* sp. n., the 82–89 pegs produce a total number of 9–29 impulses lasting for 30–70 ms in the first syllables from a group ("A" type) and 10–19 impulses lasting for 960–1770 ms in the last syllable ("B" type). In the species that stridulate ungrouped syllables: 65–85 pegs in *I. ciucasi* produce 13–56 impulses lasting for 209–438 ms, 50–80 pegs in *I. camptoxypha* produce 10–39 impulses, total duration of 27–363 ms, 48–60 pegs in *I. sicula* produce a total number of 1–8 impulses lasting for 52–265 ms. In *Isophya nagyi*, the high number of teeth on the stridulatory file, i.e. 102–109, produce a song of 36–108 impulses lasting for 167–793 ms (Table 2). All these differences in syllable production time are caused by slower or faster species–specific wing movements.

Female response as male acceptance is formed of isolated impulses, always produced after a particular part of male song, which supposedly acts as trigger. In both *Isophya dochia* sp. n. and *I. harzi* (n=13 responses from 3 ♀♀), the female replies only after the male's "B" type syllable. The same behavior was noticed in females from *I. posthumoidalis* and *I. beybienkoi* (Orci et al. 2001, Orci et al. 2010a). Females of *Isophya camptoxypha*, *I. ciucasi* and *I. sicula* reply right after the male's syllable main part. In *Isophya camptoxypha*, female response is produced immediately after the male's after–click (n=19 responses from 3 ♀♀), while in *I. ciucasi* (n=37 responses from 6 ♀♀) and *I. sicula* (n=10 responses from 1 ♀) its answer is apparently not affected by male after–clicks. Females of *Isophya nagyi* stridulate only after the last component of the male syllable (n=25 responses from 4 ♀♀) (Figs 38–43).

Table 2. Examined male song characters in close related *Isophya* species (for abbreviations see Appendix 1: *Isophya* song abbreviations) n – number of analyzed specimens; t – temperature °C.

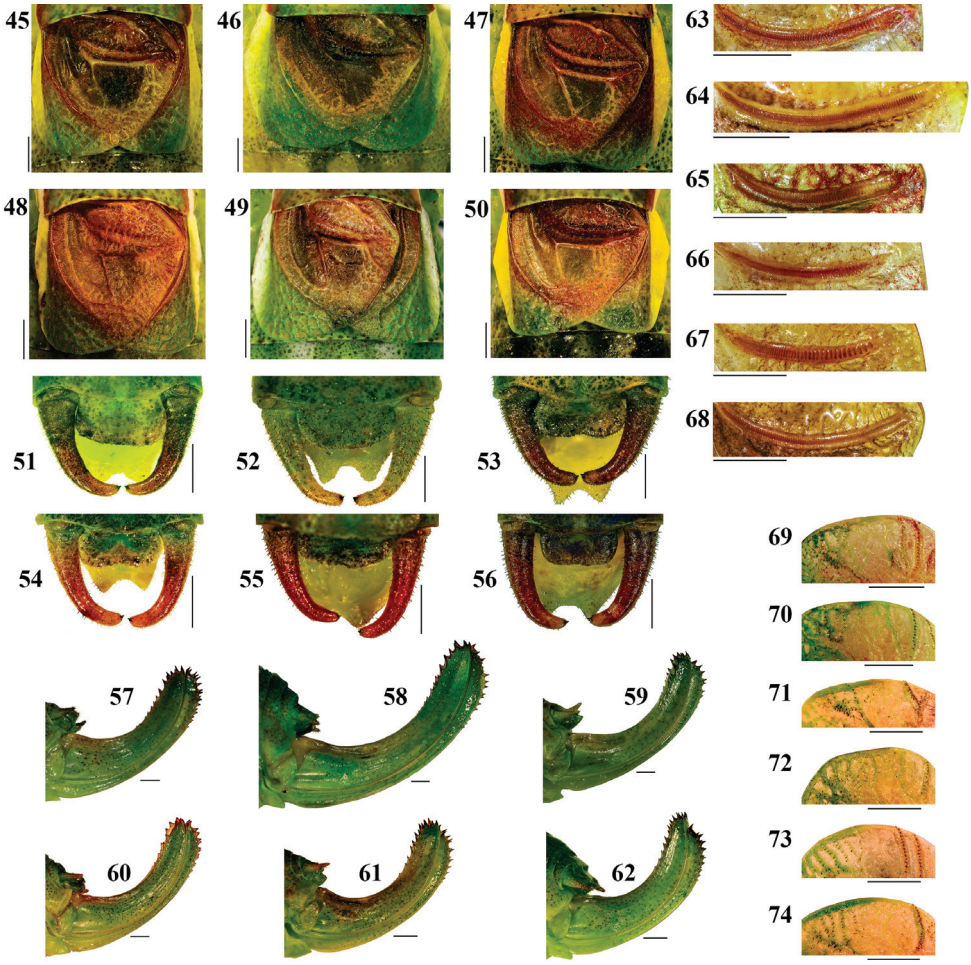
Song type	Species	Song characters	Min.	Max.	Mean	S. D.	n	t
Grouped syllable sequences	Isophya dochia sp. n.	ASD (ms)	30	70	50.74	12.83	30	20–25
		NIA	9	29	18.6	6.31		
		GS (ms)	60	131	96.28	24.62		
		BSD (ms)	30	59	39.3	9.4		
		NIB	9	15	12.36	2.1		
		DACB (ms)	621	1655	1109.1	212.65		
		NACB	1–4					
		BSTD (ms)	621	1770	1114.46	325.45		
		SGTD (ms)	1288	4761	2633.56	792.48		
		SGD (ms)	612	3028	1406.86	598.95		
		GSG (ms)	157	326	225.2	52.66		
	Isophya harzi	ASD (ms)	75	168	121.64	28.36	16	19–26
		NIA	23	55	40.5	9.67		
		DACA (ms)	40	90	64	16.97		
		NACA	0–2					
		ASTD (ms)	75	258	156.42	77.01		
		GAA (ms)	122	298	175.78	48.25		
		GAB (ms)	367	789	527.4	126.18		
		BSD (ms)	94	216	154.7	43.54		
		NIB	37	59	46.5	8.38		
		DACB (ms)	75	202	142.3	47.19		
		NACB	0–2					
		BSTD (ms)	94	418	223.3	114.78		
		SGTD (ms)	617	6168	2254.8	1954.04		
		SGD (ms)	617	6091	2126	2001.43		
		GSG (ms)	191	303	249.8	40.28		
Ungrouped syllable sequences	Isophya camptoxypha	SD (ms)	27	90	61.76	21.37	58	19–27
		NI	10	32	19.9	7.62		
		GS (ms)	110	289	189	50.18		
		DAC (ms)	56	257	112.6	70.71		
		NAC	0–1 (rarely 2–7)					
		STD (ms)	27	363	77.8	97.58		
	Isophya ciucasi	SD (ms)	7	24	15.56	5.12	34	24–28
		NI	3	26	15.32	6.74		
		GS (ms)	134	600	378.08	116.95		
		DAC (ms)	52	167	101.2	38.31		
		NAC	10–30					

Song type	Species	Song characters	Min.	Max.	Mean	S. D.	n	t
		STD (ms)	209	438	273.76	56.76		
	Isophya nagyi	SD (ms)	167	793	486.28	200.49	12	21–25
		SDC1 (ms)	9	31	21	8.08		
		NIC1	5	18	12	4.72		
		GC12 (ms)	68	301	187.6	101.94		
		SDC2 (ms)	127	221	173.9	31.57		
		NIC2	35	79	57	13.5		
		GC23 (ms)	39	97	67.5	19.04		
		SDC3 (ms)	1	35	16.4	12.33		
		NIC3	1	11	5.1	3.36		
		GS (ms)	59	235	154.26	49.59		
	Isophya sicula	SD (ms)	1	4	1.6	1.03	3	25
		NI	1	3	1.3	0.65		
		GS (ms)	190	446	337.78	62.64		
		DAC (ms)	51	110	73.2	19.71		
		NAC	0–1 (rarely 2–5)					
		STD (ms)	52	265	75.1	63.96		

Spectrographic analysis of sound reveals that in all six species the frequency ranges somewhere within interval 10–40 kHz, the maximum being recorded at about 20–30 kHz (Fig. 44).

In the six related species, males of *Isophya harzi* and *I. sicula* can be easily separated morphologically from *I. camptoxypha*, *I. ciucasi, I. nagyi* and *I. dochia* sp. n. *Isophya harzi* is more massive, males having shorter wings, marginal angle of tegmina less obtuse (110°), number of stridulatory teeth larger, and females having a longer ovipositor (11–13 mm). Males of *I. sicula* have a narrow left wing, similar with *I. posthumoidalis* and *I. beybienkoi* (Heller et al. 2004). In the other four species, the angle of cubital veins on male wing may be used as a differentiation tool: 80–90° in *I. ciucasi*, 70–80° in *I. nagyi*, 60–70° in *Isophya camptoxypha* and 70° in *I. dochia* sp. n. The tegmen marginal angle is almost constant in these species (about 120°), but less obtuse in *I. harzi* (110°) (Figs 45–50). In the six species, male cercus morphology has minute variations (Figs 51–56). The ovipositor is relatively similar in length in all species, being longer in *Isophya harzi* (Figs 57–62) and *I. beybienkoi* (Heller et al. 2004). Female stridulatory area is subject to minor intra– and interspecific variability (Figs 69–74).

Having a look at species distribution in the Romanian Carpathians, *Isophya camptoxypha* has the widest spread, inhabiting many Carpathian highlands. *Isophya harzi* is known only from two isolated mountains in the Southern Carpathians and most probably its distribution area is wider in the mountains between Prahova and Olt rivers. At this moment, both *Isophya ciucasi* and *I. dochia* sp. n. are known to have very restricted distribution areas: the isolated Ciucaş and, respectively, Ceahlău Mountain Massifs.



Figures 45–74. Morphological differences in close–related *Isophya* species: **45, 51, 57, 63, 69** *Isophya dochia* sp. n. **46, 52, 58, 64, 70** *Isophya harzi*, Piatra Craiului Mts. **47, 53, 59, 65, 71** *Isophya camptoxypha*, Nemira Mts. **48, 54, 60, 66, 72** *Isophya ciucasi*, Ciucaş Mts. **49, 55, 61, 67, 73** *Isophya sicula*, Harghita Mts. **50, 56, 62, 68, 74** *Isophya nagyi*, Călimani Mts. (**45–50** male tegmina **51–56** male cerci **57–62** ovipositor **63–68** male stridulatory file **69–74** female stridulatory bristles). Scale 1 mm.

Two species have been recently described from the Transylvanian volcanic mountains, in the Western part of the Eastern Carpathians. *Isophya nagyi* occurs in Călimani Mountains and Dorna Basin, while *I. sicula*, described from Harghita–Ciceu Mountains, has been recently found in the Moldavian Subcarpathians (Fig. 75). *Isophya posthumoidalis* was recorded in Romania only in Maramureş Basin (Szövényi and Orci 2008) and *I. beybienkoi* is known only from the area where it was described: Zadiělská planina and Plešivecká planina, Slovenskie Kras, SE Slovakia (Orci et al. 2001, Heller et al. 2004). In order to presume the evolutionary pattern within this group of species, some interesting areas must be taken into account, namely the regions where species

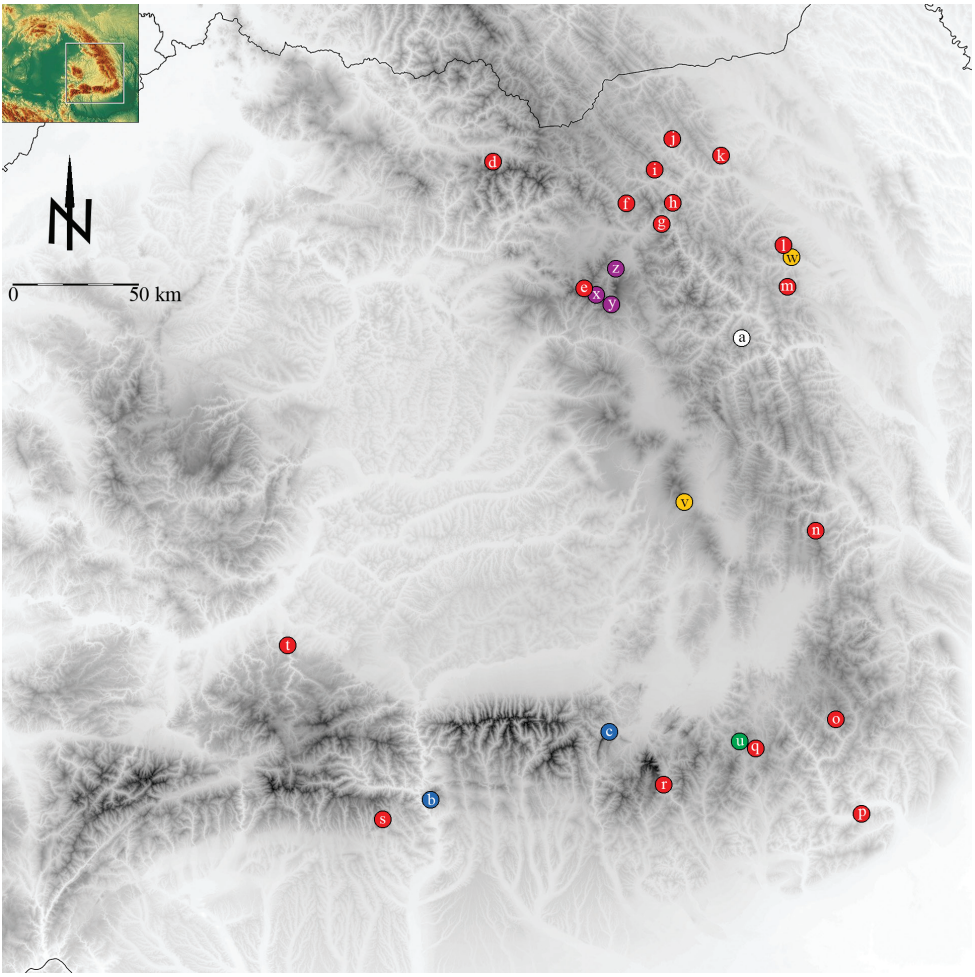


Figure 75. Distribution map of *Isophya camptoxypha* and allied species in the Romanian Carpathians, based only on acoustic analysis: white – *Isophya dochia* sp. n. (**a**); blue – *Isophya harzi* (**b, c**); red – *Isophya camptoxypha* (**d, e, f, g, h, i, j, k, l, m, n, o, p, q, r, s, t**); green – *Isophya ciucasi* (**u**); yellow – *Isophya sicula* (**v, w**); purple – *Isophya nagyi* (**x, y, z**). (**a** Ceahlău Mts. **b** Cozia Mts. **c** Piatra Craiului Mts. **d** Rodnei Mts. **e** Călimani Mts., Pietrosul peak **f** Mestecăniş **g** Pietrosul Bistriţei **h** Rarău **i** Sadova **j** Moldoviţa **k** Pleşa **l** Vânători Neamţ **m** Sihla **n** Nemira Mts. **o** Penteleu Mts. **p** Ciuta **q** Muntele Roşu, Ciucaş Mts. **r** Bucegi Mts. **s** Buila–Vânturariţa Mts. **t** Căpâlna **u** Ciucaş Mountains **v** Harghita–Ciceu Mts. **w** Vânători Neamţ **x** Călimani Mts., Pietrosul peak **y** Călimani Mts., Iezerul Călimanului peak **z** Neagra Şarului).

live syntopically: *Isophya camptoxypha* and *I. nagyi* have been found together in W Călimani Mountains, *I. camptoxypha* and *I. sicula* in Moldavian Subcarpathians, *I. camptoxypha* and *I. ciucasi* in Central Ciucaş Mountains.

The Orthoptera species of the Carpathians have been well investigated and morphologically characterized. Yet, *Isophya*, and particularly *I. camptoxypha* and its al-

lies, remain among the most intriguing and widely distributed bush–crickets in these mountains. Due to *Isophya camptoxypha*'s high intra– and interpopulational morphological variability, the main tool to separate correctly possible new cryptic species remains the oscillographic analysis of acoustic signals.

Acknowledgements

The author is grateful to Klaus–Gerhard Heller for his valuable support with literature, to Elena Iulia Iorgu, Adrian Derscariu, Liviu Moscaliuc and Irinel Popescu, his traveling companions in the Carpathians and to Lucian Fusu for inspiration during the preparation of manuscript. Ceahlău National Park, Rodnei National Park and Călimani National Park Administrations are acknowledged for the excellent work conditions. Advices of anonymous reviewers significantly improved the paper.

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Appendix 1

Isophya song abbreviations. (doi: 10.3897/zookeys.254.3892.app1) File format: Microsoft Excel file (xls).

Explanation note: Abbreviations used for calling songs of *Isophya* males, described in Figures 1-4 and Table 2.

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Appendix 2

Isophya dochia sp. n. audio. (doi: 10.3897/zookeys.254.3892.app2) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya dochia* sp. n.

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Appendix 3

Isophya dochia sp. n. video. (doi: 10.3897/zookeys.254.3892.app3) File format: Video file (MPEG Layer 4) (mp4).

Explanation note: Video recording of calling song of a male *Isophya dochia* sp. n.

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Appendix 4

Isophya comparative material. (doi: 10.3897/zookeys.254.3892.app4) File format: Microsoft Excel file (xls).

Explanation note: Acoustic analyzed *Isophya* material in the Romanian Carpathian Mountains.

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Appendix 5

Isophya harzi audio. (doi: 10.3897/zookeys.254.3892.app5) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya harzi*.

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Appendix 6

Isophya camptoxypha audio. (doi: 10.3897/zookeys.254.3892.app6) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya camptoxypha*.

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Appendix 7

Isophya ciucasi audio. (doi: 10.3897/zookeys.254.3892.app7) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya ciucasi*.

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Appendix 8

Isophya sicula audio. (doi: 10.3897/zookeys.254.3892.app8) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya sicula*.

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Appendix 9

Isophya nagyi audio. (doi: 10.3897/zookeys.254.3892.app9) File format: Windows audio file (wav).

Explanation note: Audio recording of calling song of a male *Isophya nagyi*.

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ZooKeys 254: 23–45 (2012)
doi: 10.3897/zookeys.254.3959
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RESEARCH ARTICLE

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Morphological and molecular taxonomy of *Nidularia balachowskii* Bodenheimer (Hemiptera, Coccoidea, Kermesidae) with notes on its life history in Israel

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Academic editor: *M. Wilson* | Received 6 September 2012 | Accepted 23 November 2012 | Published 21 December 2012

Citation: Spodek M, Ben-Dov Y, Ghanim M, Mendel Z (2012) Morphological and molecular taxonomy of *Nidularia balachowskii* Bodenheimer (Hemiptera, Coccoidea, Kermesidae) with notes on its life history in Israel. ZooKeys 254: 23–45. http://dx.doi.org/10.3897/zookeys.254.3959

Abstract

Descriptions and illustrations of the adult female and first-instar nymph of the kermesid *Nidularia balachowskii* Bodenheimer, based on the adult female lectotype and paralectotype (here designated), and new material collected from Israel are presented. A key for the identification of first-instar nymphs of *Nidularia* spp. is offered. Molecular identification of *N. balachowskii*, using nucleotide sequences from the D2–D3 region of the 28S ribosomal gene, and the mitochondrial Cytochrome Oxidase I (COI) gene, is presented. Morphological and molecular analyses confirm that *N. balachowskii* is closely related to other species within the Kermesidae. In Israel, this species develops only on *Quercus ithaburensis* and is univoltine. This is the first detailed report of *N. balachowskii* from Israel.

Keywords

Scale insect, *Quercus* spp., morphology, univoltine, monophagous, 28S, COI

Introduction

Nidularia Targioni-Tozzetti, 1868 is one of ten genera of scale insects within the Kermesidae (Hemiptera: Coccoidea). Three species of *Nidularia*: *N. balachowskii* Bodenheimer, 1941, *N. japonica* Kuwana, 1918 and *N. pulvinata* (Planchon, 1864),

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have been recorded so far only from the Palaearctic region (Ben-Dov et al. 2012). *Nidularia pulvinata,* the type species of this genus (Signoret 1875, Morrison and Morrison 1966), occurs in western territories of the Palaearctic region, where it has been collected on three oak species, *Quercus coccifera* L., *Q. ilex* L. and *Q. ithaburensis* Decne. (Koteja 1980, Bullington and Kosztarab 1985).

The three species of *Nidularia* have been recorded as follows: *N. pulvinata* from Algeria (Koteja 1980); China (Tang 1984); France, including Corsica (Balachowsky 1933, Foldi 2001, 2003); Italy (Hoy 1963, Viggiani 1991); Portugal (Hoy 1963); Spain (Gomez-Menor Ortega 1937, Hoy 1963); *N. japonica* in China and Japan, on the branches and trunks of several oak species namely, *Quercus acutissima* Carruth., *Q. ailena* Blume, *Q. dentate* Thunb., *Q. fabri* Hance., *Q. glandulfera* Blume (Liu et al. 1997); while *N. balachowskii* has been reported from Turkey (Bodenheimer 1941), Iran and Israel (Bodenheimer 1944) on *Quercus* sp..

The family placement of *Nidularia* has changed over the years. Targioni-Tozzetti (1868) placed this genus in the Coccidae *sensu lato.* Signoret (1875) and Koteja (1974, 1980) restricted it to the Eriococcidae. Currently, it is accepted that this genus belongs to the Kermesidae (Morrison and Morrison 1966, Bullington and Kosztarab 1985, Baer and Kosztarab 1985, Ben-Dov et al. 2012).

Most species of the Kermesidae appear to be univoltine (Balachowsky 1950, 1953, McConnell and Davidson 1959, Hamon et al. 1976, Sternlicht 1969, Koteja 1980, Bullington and Kosztarab 1985, Hu 1986, Kosztarab and Kozár 1988, Viggiani 1991, Liu et al. 1997, Marotta et al. 1999). Females develop through three nymphal instars before reaching maturity (Bullington and Kosztarab 1985). Males are only known for a few species and the remaining species are thought to reproduce parthenogenetically (McConnell and Davidson 1959, Sternlicht 1969, Koteja and Zak-Ogaza 1972, Hamon et al. 1976, Baer and Kosztarab 1985, Hu 1986, Kosztarab and Kozár 1988, Miller and Miller 1993, Liu et al. 1997, Marotta et al. 1999, Turner 2004). Kermesidae are oviparous and, after the female's last molt and before oviposition, the scale's body increases its size and the dorsum becomes convex and sclerotized. The female lays eggs in the brood chamber which is located beneath the female's venter (Bullington and Kosztarab 1985). The size, shape, and color pattern of post-reproductive females varies considerably within the same species (Bullington and Kosztarb 1985).

The body of post-reproducing females may remain on the host tree for a year or more after the emergence of the first-instar nymphs (Baer 1980, authors' observations). Most species of Kermesidae are not known to cause any visible injury to their host trees although there are reports of branch dieback, flagging, reduced growth rates and occasional tree death (Kozár 1974, Hamon 1977, Solomon et al. 1980, Viggiani 1991, Pellizzari et al. 2012). On the other hand, some species of *Kermes* are known for their importance as a source of crimson dye (Amar 2005, Cardon 2007).

The main synapomorphic characters of kermesid adult females have been summarized by several authors including: Ferris (1955), Bullington and Kosztarab (1985), Hodgson (1997) and Miller et al. (2005). They are the presence of: (i) bilocular pores on venter; (ii) simple pores on dorsum; (iii) tubular ducts; (iv) a three-segmented la-

bium with setae; (v) a group of multilocular disc-pores near the base of each antenna; (vi) an anal ring with or without cells and (vii) anal ring with or without setae and absence of: (viii) a spiracular band of quinquelocular disc-pores, (ix) stigmatic spines and (x) ventral microducts.

Species of *Nidularia* share morphological and biological characteristics with other species of the Kermesidae. Like *Kermes* spp., *Nidularia* spp. are monophagous and develop on oak trees. They are known as 'gall-like insects' due to the size and body shape of the convex and sclerotized post-reproductive adult female (Kosztarab and Kozár 1988). They mainly develop in bark crevices, forks between small twigs and buds, and on branches (Bullington and Kosztarab 1985).

Seven species of Kermesidae belonging to two genera, *Kermes* Boitard and *Nidularia* Targioni-Tozzetti, are recorded from Israel (Ben-Dov et al. 2012). Two species, namely *K. greeni* and *K. nahalali*, were originally described from post-reproductive adult females (Bodenheimer 1931), three species: *K. echinatus, K. palestiniensis* and *K. spatulatus* were described from the first-instar nymphs (Balachowsky 1953), and one species, *K. bytinskii*, was described from the adult female and all nymphal instars by Sternlicht (1969). *Nidularia balachowskii* was originally described from Turkey (Bodenheimer 1941), and then, in the same year, it was collected in northern Israel at Daphne Oaks, which is today known as Horshat Tal Nature Reserve. Until now, the material collected from Daphne Oaks was the only known record of *N. balachowskii* in Israel.

Between 2010 and 2012, we surveyed the scale insect fauna of various species of oak trees in Israel. *N. balachowskii* was found on branches of *Q. ithaburensis* over a wide range of oak forests in northern Israel. Although Hoy (1963) and Koteja (1980) recorded *N. pulvinata* from Israel, we did not encounter this species during our forest surveys. In addition, no material of *N. pulvinata* from Israel was found among the Kermesidae records of dry material and microscope slides collections examined at the MNHN, BMNH, TAU and ICVI. To date it seems that *N. balachowskii* is the sole *Nidularia* species present in Israel.

Bodenheimer's original description of *N. balachowskii* contains a description and some illustrations of major characters in the adult female (Bodenheimer 1941). His description is short and incomplete. The main objective of this paper is to redescribe *N. balachowskii* and to establish its generic and family placement. Our descriptions and illustrations are based on the lectotype and paralectotype specimens (see Material examined) as well as on fresh material collected from Israel and Turkey. In addition, the first-instar nymph is described and illustrated for the first time. An identification key for the first-instar nymphs of *Nidularia* is provided. We also compare some characters of the adult female of *N. balachowskii* with those of several other species of Kermesidae: *N. pulvinata, Kermes roboris* (Fourcroy) (type species of *Kermes*) and *K. quercus* (Linnaeus). In addition, sequences of 28S and COI genes from *N. balachowskii* and five or six other kermesids species plus some scale insect representatives of other Coccoidea families were compared in order to elucidate the family placement *of N. balachowskii.* Information on habitat and life history of *N. balachowskii* in Israel is also provided.

Material and methods

Specimen collections

This redescription of *N. balachowskii* is based on type material (see Material examined below), plus specimens from Israel collected by Bodenheimer and fresh material collected in Israel by us. Populations of *N. balachowskii* from *Q. ithaburensis* trees were studied and specimens were collected between 2010 and 2012 from the following nature reserves in northern Israel: Yehudiya Nature Reserve, Golan Heights (32°56'19"N, 35°39'56"E); Horshat Tal Nature Reserve, Upper Galilee (33°13'13.74"N, 35°37'45.65"E); Alonei Abba Nature Reserve, Lower Galilee (32°43'46.2"N, 35°10'18.47"E). Trees at each reserve were surveyed at least once a month and 150–200 branches (20–25 cm in length) were removed at each visit. The branches were taken back to the laboratory in large plastic bags and examined individually under a stereomicroscope for scale insects. Relevant specimens were slide-mounted for microscope examination using the protocol in Ben-Dov, Hodgson (1997). Specimens of *N. pulvinata* (adult females and first-instar nymphs) became available from MNHN. Material of *N. japonica* was not available and comparisons with this species were based on the original description by Kuwana (1918) as well on the redescription by Liu et al. (1997). Dry and mounted material of *N. balachowskii* from Israel, are deposited in the ICVI, BMNH and MNHN.

Identification and morphological observations

Illustrations of the adult female and the first-instar nymph of *N. balachowskii* are generalizations of several specimens, showing the dorsum on the left and the venter on the right, with enlargements of important structures arranged around the main drawing. The enlarged structures are not drawn to the same scale. Terms for morphological features follow chiefly those of Bullington and Kosztarab (1985), Baer and Kosztarab (1985) and Hodgson (1994). Measurements of specimens and of morphological structures were made using an ocular micrometer on an Olympus BX51 phase contrast microscope. Measurements of structures are given in microns (μm) and millimeters (mm). Body length was measured from the farthest points of the head to the posterior end of the body, and body width was the greatest width. Setae lengths were measured from the tip of its base (excluding the setal socket) to the apical tip of the setae. The frequency of each structure is given for the entire body. The range is taken from twenty specimens.

Abbreviations of specimen depositories are as follows: **BMNH** - The Natural History Museum, London, U.K.; **ICVI** - Coccoidea Collection, Department of Entomology, Agricultural Research Organization, Bet Dagan, Israel; **MNHN** - Museum National d' Histoire Naturelle, Paris, France; and **TAU** - Tel Aviv University Insect Collection, Israel.

Material examined

Nidularia balachowskii

Turkey: **Lectotype** female (ICVI), here designated, and **paralectotype** female (MNHN), 21 km at road from Mardin to Diyarbakir, on branches and twigs of *Quercus* sp. (Fagaceae), 13.ii.1939, F.S. Bodenheimer. Bodenheimer (1941) did not select a holotype, and we regard the above-mentioned specimens as the original material studied by him as indicated on the slide labels.

Additional non-type material from **Turkey** as follows: Van-Koçet Road (alt.1625 m) on *Quercus* sp., 19.vii.2005, B. Kaydan (Yuzuncu Yil University, Turkey 2056); Hakkari -Üzümcü Road (alt. 956 m) on *Quercus* sp., 15.ix.2005, B. Kaydan (Yuzuncu Yil Universty, Turkey 2343); Van-Hakkari Road (alt. 1266 m) on *Quercus* sp., 16.ix.2005, B. Kaydan (Yuzuncu Yil Universty, Turkey 2370); Hakkari-Doğan (alt. 1032 m) on *Quercus* sp., 22.v.2005, B. Kaydan (Yuzuncu Yil Universty, Turkey 2688); Bitlis River (alt. 797 m) on *Quercus sp.*, 23.vi.2006, B. Kaydan (Yuzuncu Yil Universty, Turkey 3036); Bitlis-Kavakbaşı (alt. 1365) on *Quercus* sp. 30.v.2007, B. Kaydan (Yuzuncu Yil Universty, Turkey 3419).

Israel: adult female, Daphne Oaks (= current name Horshat Tal Nature Reserve), on *Quercus* sp., 1.v.1939, F.S. Bodenheimer, (ICVI C:4805). This was the first record of this species from Israel. Additional females and first-instar nymphs, all collected off *Q. ithaburensis* by M. Spodek: Yehudiya Nature Reserve, 10.x.2010, 7.xi.2010, 11.i.2011, 6.ii.2011, 16.x.2011, 6.xi.2011, 3.vi.2012, (ICVI C:4891, C:4912, C:4945, C:4970, MC:587, C:4970, MC:690); Horshat Tal Nature Reserve, 30.v.2010, 14.ii.2012, 27.ii.2011, 13.iii. 2012, (ICVI MC:228, MC:614, MC:430, C:5131); Alonei Abba Nature Reserve, 11.1.2011, (ICVI MC:385). First-instar nymphs; Yehudiya Nature Reserve, 6.iii.2010, 20.iii.2011, 24.iii.2012, (ICVI MC:140, MC:460, MC:637); Horshat Tal Nature Reserve, 13.iii.2012, 15.iii.2012, (ICVI MC:635, MC:636).

Comparative material examined

Nidularia pulvinata: **France**: adult female and first-instar nymphs, Serignan, Vauctuse, on *Q. ilex,* 18.v.1978, I. Foldi (ICVI C:4946); first-instar nymphs, Caumont (Avignon), on *Q. ilex*, 11.iv.1978, D. Matile, J.P. Fabre, (MNHN 7337-4); **Italy**: adult female Pisa, on *Q. ilex*, 5.iv.1988, D. Matile-Ferrero (MNHN10959, ICVI 5060)

Kermes quercus: **Sweden**: adult female, Skan, Near Lund on *Q. robur,* 10.vi.2010, C.A Gertsson (ICVI C:4806); **England**: adult female , Wytham Wood, Berkshire on *Q. robur,* 10.v.1965, S.C. Varley (BMNH 81-539); **Poland**; first-instar nymph, Warsaw on *Q. robur*, 25.viii.1994, E. Podsiadlo (ICVI C:4798)

Kermes roboris: **Hungary**: adult female, Budapest, Plant Protection Institute-adjacent track on *Quercus* sp., 8.vi.1989, C.P. Malumphy (BMNH); **England**: adult female, Kent: Herne Bay on *Quercus* spp., 00.viii.1899, C.D Waterhouse (ICIV C:5071).

Molecular methods

Samples

Specimens of the following adult female kermesid species, identified by YBD, were used in the molecular part of this study: *K. nahalali* Bodenheimer, *K. echinatus* Balachowsky, *K. greeni* Bodenheimer, *K. quercus* (Linnaeus), *K. spatulatus* Balachowsky and *N. balachowskii*. Three specimens of each species were used as replicates except for *N. balachowskii*, where six were used. Adult females, preserved in 96% ethanol, were examined under the stereomicroscope for the presence of hymenopteran parasitoid wasps prior to DNA extraction. Voucher specimens were slide-mounted using the cuticle of the actual specimens from which DNA was extracted. Slide mounting followed the protocol outlined in Ben-Dov, Hodgson (1997), and the voucher slides are deposited in the ICVI (Table 1). To provide some taxonomic context to our study, we included also DNA sequences of species belonging to other families within the Coccoidea: Asterolecaniidae, Coccidae, Diaspididae, Eriococcidae, Monophlebidae and Pseudococcidae. A species of aphid, *Acyrthosiphon pisum* (Hemiptera, Aphidoidea), was used as the outgroup species. These sequences were made available from GenBank (Table 1). The sequences of Kermesidae species obtained in this study are deposited in the GenBank under the accession numbers JX436113 - JX436154.

DNA extraction, amplification and sequencing

DNA was extracted from parasitoid-free adult females using the Cetyl trimethylammonium bromide (CTAB) method (Murray and Thompson 1980). Polymerase chain reaction (PCR) products were generated from the mitochondrial Cytochrome Oxidase I (COI) gene, and a fragment of the D2 and D3 regions of the 28S ribosomal DNA gene. PCR reaction was performed in a total volume of 25 µL containing 1 unit of dream Taq polymerase (Fermentas, USA), 2.5 µL of enzyme buffer supplemented with MgCl2, 0.2 µL of 25 mM dNTPs, 0.3 µL of 20 pmole for each primer, and 2 µL of DNA template. A 900 bp fragment of the 28S ribosomal RNA gene and a 400 bp fragment of the COI gene were amplified and sequenced. Primers for both genes were 28S forward 5'-GAC CCG TCT TGA AAC ACG GA-3' and 28S reverse 5'-TCG GAA GGA ACC AGC TAC TA-3' (Gullan et al. 2010). COI forward 5'-CAA CAT TTA TTT TGA TTT TTT GG-3' (C1-J-2183 aka Jerry) and COI reverse 5'-GCW ACW ACR TAT AKG TAT CAT G-3' (C1-N-2568 aka Ben) (Gullan et al. 2010). The COI barcode region (Herbert et al. 2003) was not used because it has failed to amplify in most scale insects tried to date (Schroer et al. 2008).

The PCR cycling conditions for 28S were 94°C for 4 min, followed by 35 cycles of 94°C for 1 min, 50°C for 1 min, and 72°C for 1.5 min, with a final extension at 72°C for 4 min. The PCR cycling protocol for COI was 95°C for 7 min, followed

Table 1. Collection information and GenBank accession numbers for insect samples used in this study.

Species name	Family	Voucher code	Host tree	Location collected	Date collected	Collector	GenBank Accession No. 28S	GenBank Accession No. COI
Kermes nahalali Bodenheimer	Kermesidae	C-5111	*Quercus ithaburensis*	ISRAEL: Alonei Abba Reserve	27.ii.2011	M. Spodek	JX436134	JX436113
Kermes nahalali Bodenheimer	Kermesidae	C-5112	*Quercus ithaburensis*	ISRAEL: Alonei Abba Reserve	27.ii.2011	M. Spodek	JX436135	JX436114
Kermes nahalali Bodenheimer	Kermesidae	C-5113	*Quercus ithaburensis*	ISRAEL: Alonei Abba Reserve	27.ii.2011	M. Spodek	JX436136	JX436115
Kermes echinatus Balachowsky	Kermesidae	C-5114	*Quercus calliprinos*	ISRAEL: Alonei Abba Reserve	19.vi.2011	M. Spodek	JX436137	JX436116
Kermes echinatus Balachowsky	Kermesidae	C-5115	*Quercus calliprinos*	ISRAEL: Alonei Abba Reserve	19.vi.2011	M. Spodek	JX436138	JX436117
Kermes echinatus Balachowsky	Kermesidae	C-5116	*Quercus calliprinos*	ISRAEL: Alonei Abba Reserve	19.vi.2011	M. Spodek	JX436139	JX436118
Kermes greeni Bodenheimer	Kermesidae	C-5117	*Quercus calliprinos*	ISRAEL: Hanita	8.vi.2011	M. Spodek	JX436140	JX436119
Kermes greeni Bodenheimer	Kermesidae	C-5118	*Quercus calliprinos*	ISRAEL: Hanita	8.vi.2011	M. Spodek	JX436141	JX436120
Kermes greeni Bodenheimer	Kermesidae	C-5119	*Quercus calliprinos*	ISRAEL: Hanita	8.vi.2011	M. Spodek	JX436142	JX436121
Kermes quercus (Linnaeus)	Kermesidae	C-5120	*Quercus robur*	SWEDEN: Skan, near Lund	10.vi.2010	C. A. Gertsson	JX436143	JX436122
Kermes quercus (Linnaeus)	Kermesidae	C-5121	*Quercus robur*	SWEDEN: Skan, near Lund	10.vi.2010	C. A. Gertsson	JX436144	JX436123
Kermes quercus (Linnaeus)	Kermesidae	C-5122	*Quercus robur*	SWEDEN: Skan, near Lund	10.vi.2010	C. A. Gertsson	JX436145	JX436124
Kermes spatulatus Balachowsky	Kermesidae	C-5123	*Quercus ithaburensis*	ISRAEL: Horshat Tal Reserve	3.iv.2011	M. Spodek	JX436146	JX436125
Kermes spatulatus Balachowsky	Kermesidae	C-5124	*Quercus ithaburensis*	ISRAEL: Horshat Tal Reserve	3.iv.2011	M. Spodek	JX436147	JX436126
Kermes spatulatus Balachowsky	Kermesidae	C-5125	*Quercus ithaburensis*	ISRAEL: Horshat Tal Reserve	3.iv.2011	M. Spodek	JX436148	JX436127

Species name	Family	Voucher code	Host tree	Location collected	Date collected	Collector	GenBank Accession No. 28S	GenBank Accession No. COI
Nidularia balachowskii Bodenheimer	Kermesidae	C-5126	*Quercus ithaburensis*	ISRAEL: Yehudiya Reserve	11.i.2011	M. Spodek	JX436149	JX436128
Nidularia balachowskii Bodenheimer	Kermesidae	C-5127	*Quercus ithaburensis*	ISRAEL: Yehudiya Reserve	11.i.2011	M. Spodek	JX436150	JX436129
Nidularia balachowskii Bodenheimer	Kermesidae	C-5128	*Quercus ithaburensis*	ISRAEL: Alonei Abba Reserve	27.ii.2011	M. Spodek	JX436151	JX436130
Nidularia balachowskii Bodenheimer	Kermesidae	C-5129	*Quercus ithaburensis*	ISRAEL: Alonei Abba Reserve	27.ii.2011	M. Spodek	JX436152	JX436131
Nidularia balachowskii Bodenheimer	Kermesidae	C-5130	*Quercus ithaburensis*	ISRAEL: Horshat Tal Reserve	14.ii.2012	M. Spodek	JX436153	JX436132
Nidularia balachowskii Bodenheimer	Kermesidae	C-5131	*Quercus ithaburensis*	ISRAEL: Horshat Tal Reserve	14.ii.2012	M. Spodek	JX436154	JX436133
Kermes nakagawae Kuwana	Kermesidae						n/a	AB439525.1
Bambusaspis miliaris (Boisduval)	Asterolecaniidae						GU998966.1	n/a
Ceroplastes rubens Maskell	Coccidae						n/a	JQ795720.1
Paralecanium sp.	Coccidae						GU 998968.1	n/a
Pelliculaspis celtis McDaniel	Diaspididae						GQ325525.1	n/a
Apiomorpha nookara Mills	Eriococcidae						n/a	JN863289.1
Eriococcus spurius (Modeer)	Eriococcidae						GU998969.1	n/a
Drosicha mangiferae (Green)	Monophlebidae						n/a	JF792882.1
Phenacoccus parvus Morrison	Pseudococcidae						n/a	JQ863289.1
Outgroup								
Acyrthosiphon pisum Harris	Aphididae						S50426.1	EU701281.1

by 40 cycles of 95°C for 1 min, 45°C for 1 min, and 72°C for 1.5 min, with a final extension at 72°C for 5 min. Each reaction was examined by electrophoresis and bands were visualized with UV light. PCR products were excised from the gel and purified using the Zymoclean Gel Extraction Kit (Zymo Research, Irvine, CA). Purified PCR products were sequenced in both the forward and reverse directions at Hy-Labs (Rehovot, Israel).

Sequence alignment and phylogenetic analysis

Sequence alignments for both 28S and COI gene sequences were performed with MUSCLE 3.7 (Edgar 2004) and the results were adjusted manually where necessary to maximize alignment. The alignment data for each gene were used in maximum likelihood tree construction, using Kimura-2 parameter model (K2P) genetic distances (Kimura 1980). Both trees were generated using MEGA v.5 (Tamura et al. 2011) and branch support was estimated with 1000 bootstrap replicates.

Results

Morphological characteristics

***Nidularia balachowskii* Bodenheimer**
http://species-id.net/wiki/Nidularia_balachowskii

Nidularia balachowskii Bodenheimer, 1941: 78–80.

Adult female. General appearance. Young, pre-reproductive adult dorsum brownish and venter yellowish white; oval, soft and flat; 1.2–1.9 mm long and 0.6–0.9 mm wide. Dorsal surface covered with 5 longitudinal rows of rectangular wax plates, each plate about 0.25 mm long and 0.3 mm wide; median row with 11 plates, lateral row on each side of median row with 9–11 plates and marginal rows with 7–9 plates. The wax plates become gradually smaller in size towards anterior and posterior apices and lateral margin (Fig. 1). **Post-reproductive female** oval, moderately convex and sclerotized; 2.75–3.75 mm long, 2–3 mm wide and 0.8–1.8 mm high; 5 longitudinal rows of dark brown wax plates almost fused; with lighter brown wax in between rows of plates (Fig. 2).

Slide-mounted young adult female 0.8– 3 mm long, 0.5–2.5 mm wide (Fig. 3).

Margin. Marginal setae, pointed, 12–13 μm long; placed in a row of 30–36 setae. Stigmatic spines absent. Anal cleft absent.

Dorsum. Simple pores, circular, each with a sclerotized rim and 1 μm diameter; covering entire dorsum. Other pore types absent. Dorsal setae absent.

Figure 1. *Nidularia balachowskii* Bodenheimer young adult female, general appearance.

Venter. Eyes circular, 5–7 µm diameter, each placed anterolaterally to each antenna. **Antennae** each 1-segmented, 15–25 µm long, 12–18 µm wide**;** each bearing 2–6 fleshy setae. **Legs** absent. **Clypeolabral shield** 113–155 µm long, 113–125 µm wide. **Labium** 3-segmented, triangular, 100–125 µm long, 50–63 µm wide; labial setae as follows; basal segment with 2 pairs of hair-like setae, 9–20 µm long, median segment with 1 pair of hair-like setae, 11–13 µm long, apical segment with 6 setae; 2 apical setae, 9–12 µm long plus 4 hair-like; subapical setae, each 12–20 µm long. **Spiracles** subequal in size; each 42–60 µm long, 31–50 µm diameter of peritreme. **Quinquelocular pores** each 5 µm diameter; with 8–11 between mesothoracic spiracles and submarginal band of tubular ducts; 10–13 between the metathoracic spiracles; 2–4 laterad to each metathoracic spiracle; also in a single, complete submarginal band, 1 pore wide from head apex to anal ring; total number of pores per side about 50–71.**Bilocular pores** each about 3 µm wide, totaling 103–135 per side, dispersed within a submarginal tubular duct band. **Multilocular pores** each 7–8 µm in diameter with 9–10 loculi; in groups of 3 or 4 between each metathoracic and mesothoracic spiracle; in transverse bands across abdominal segments arranged as follows; segment III with 3–7, IV with 12–24, V with 14–28, VI with 15–35, VII with 18–25, VIII with 12–19, IX with 2–3 on each side of vulva. **Tubular ducts**

Figure 2. *Nidularia balachowskii* Bodenheimer post-reproductive female, general appearance.

dispersed in a complete submarginal band, 2–3 ducts wide, each duct with outer ductule 12–18 µm long and 5 µm wide, a sclerotized cup about 5 µm in diameter and inner ductule about 22–30 µm long; also scattered over thorax. **Other ventral setae:** with a group of 7 or 8 setae, each 7–13 µm long, anterior to clypeus; 1 pair, 7–8 µm long, posterior to each antenna; 8–12 setae, each 5–8 µm long, placed medially to each spiracle; 8 setae, 5–13 µm long, distributed in 1 longitudinal row placed medially to each marginal band of bilocular pores and tubular ducts; each abdominal segment with transverse rows of 4–10 setae, each 7–8 µm long, placed anterior to bands of multilocular pores. **Microspines** present on median and sub-median areas of each abdominal segment, in 3–5 transverse rows; each spine about 1 µm long, also scattered on thorax. **Anal ring** located on venter, composed of 2 semi-circles; diameter 27–35 µm; each half circle bearing 3 pointed setae and 10–12 cells; anterior setae each 25–38 µm long, median and posterior setae each 15–25 µm long; 2 pairs of thin setae just anterior to anal ring, each 7–10 µm long, plus a pair of pointed setae postero-laterally to anal ring, separated by a space about double diameter of anal ring, each 10–13 µm long. Also 1 pair of apical setae, each 65–68 µm long, and 4 setae, each 15–22 µm long, between apical setae, similar in structure to, but longer than, marginal setae.

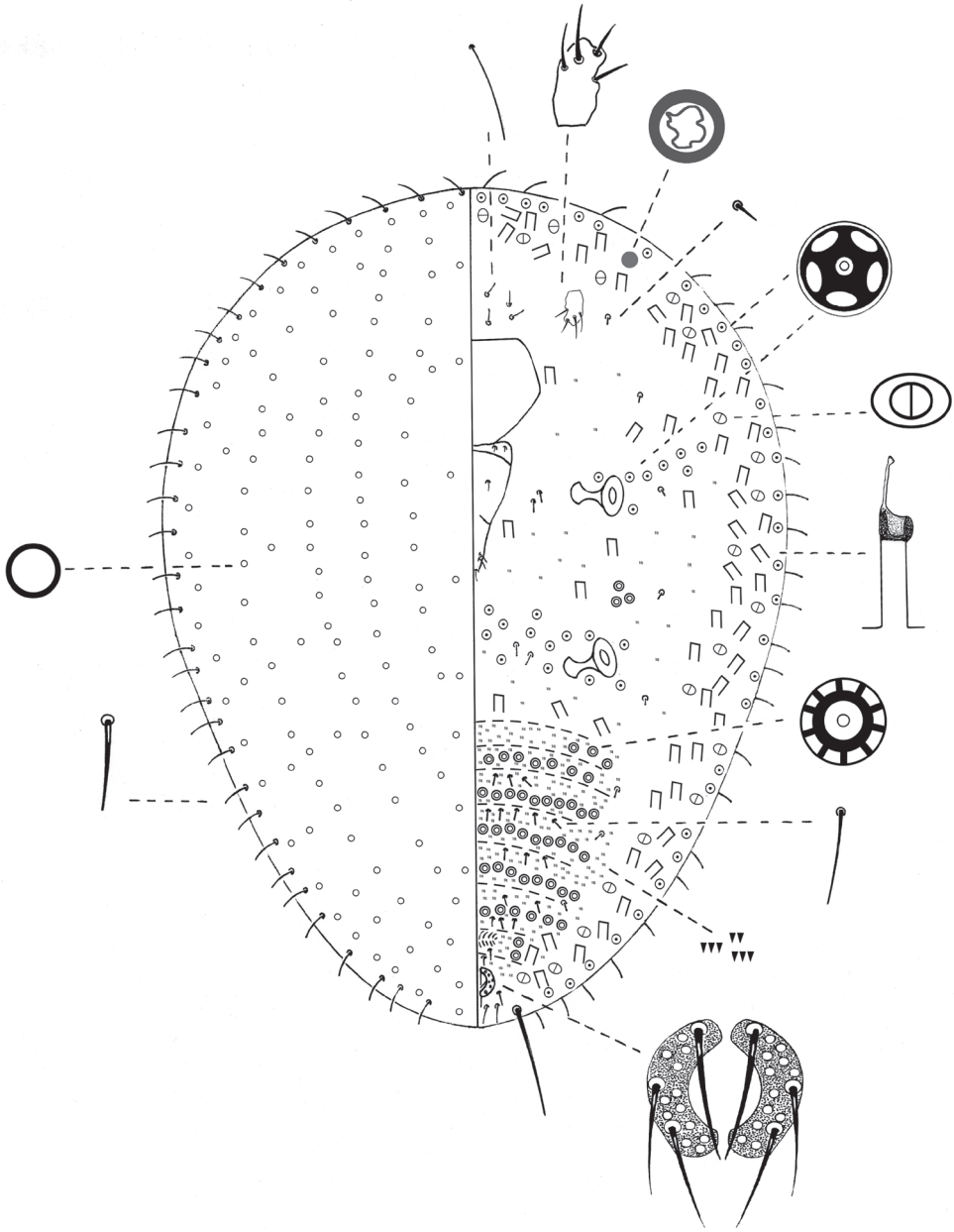

Figure 3. *Nidularia balachowskii* Bodenheimer adult female.

First-instar nymph. General appearance. Yellow-greyish, oval and tapering posteriorly 0.38–0.43 mm long and 0.2–0.3 mm wide.

Slide-mounted specimen. Oval, 0.42–0.53 mm long and 0.19–0.34 mm wide (Fig. 4)**.**

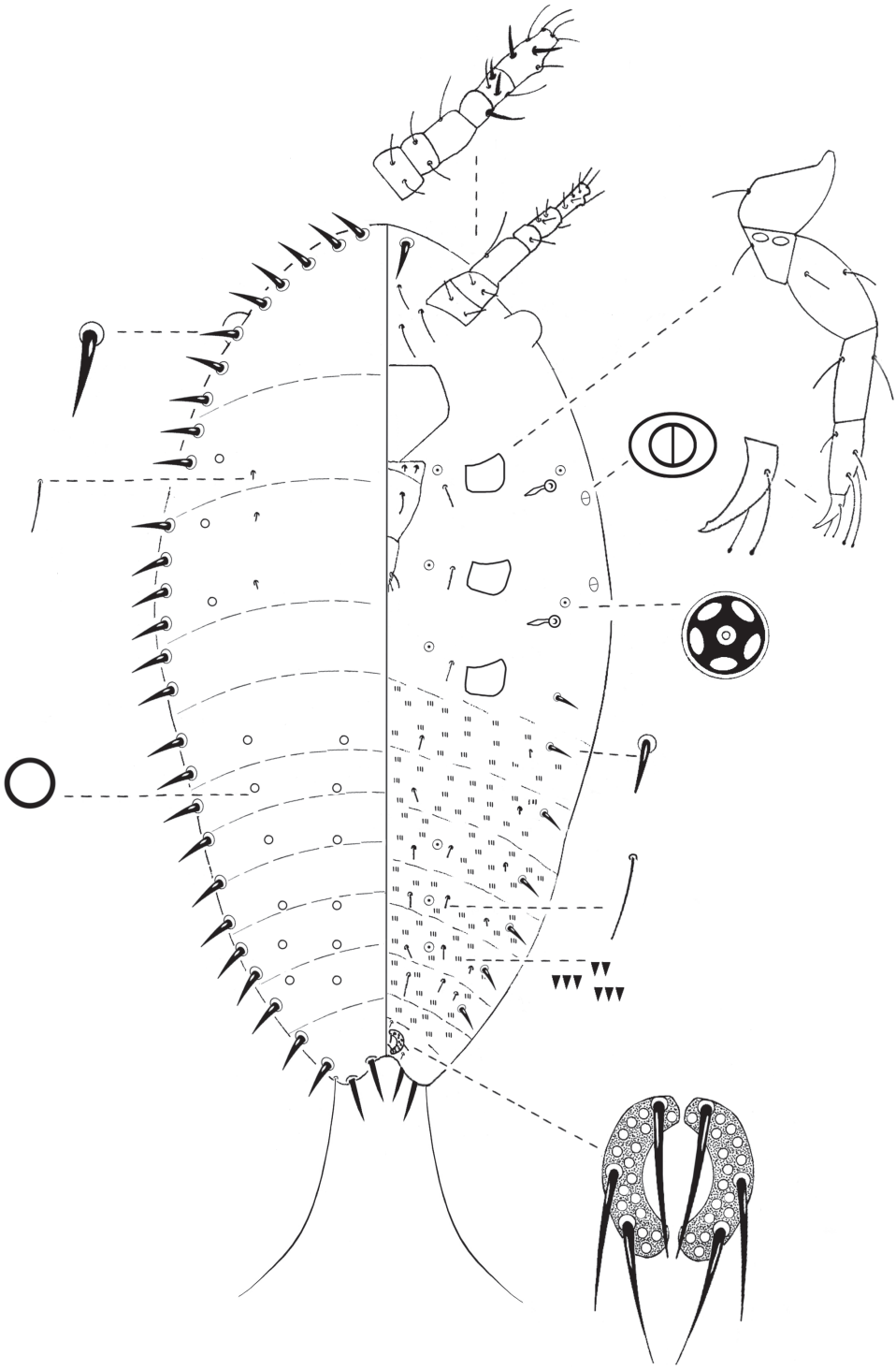

Figure 4. *Nidularia balachowskii* Bodenheimer first-instar nymph.

Margin. Marginal setae sharply spinose, pointed apically and slightly curved; each 9–15 μm long; in a distinct row of 26–33 setae on each margin.

Dorsum. Derm membranous; intersegmental lines observable. **Simple pores**, circular with a sclerotized rim, each 1 μm diameter; totaling about 30, scattered over entire dorsum in 4 longitudinal rows; 2 submarginal lines on thorax and abdomen and 2 medial lines on abdomen; with a single pair of submedial setae on each thoracic segment, each 5 μm long.

Venter. Eyes present as semi-circles near margin, diameter 12–15 μm. **Antennae** each 6-segmented, length 88–125 μm; segments III and VI longer than other segments; scape with 2 hair-like setae; pedicel with 2 hair-like setae; segment III with 1 long hair-like seta; IV with 1 fleshy seta; V with 1 fleshy seta, 2 hair-like setae and, 1 thick hair-like seta; apical segment with 2 fleshy setae and 5 hair-like setae. **Legs** well-developed; measurements of hind legs; (in μm): coxae 25–30, trochanter + femur 63–80, tibia 25–38, tarsus 25–60, claw 13–23; total leg length 158–213; trochanter with 2 oval, sensory pores on each side, each about 2–3 μm wide; setae present on each leg segment; tarsal digitules each 25–30 μm long, knobbed apically, extending beyond apex of claw; claw digitules knobbed apically, each 14–20 μm long, shorter than tarsal digitules; each claw with a single denticle near the tip. **Clypeolabral shield** well-developed; 75–90 μm long and 63–75 μm wide. **Labium** 3-segmented, triangular, 82–100 μm long and 35–38 μm wide; labial setae as follows; basal segment with 2 pairs of setae, each 10–15 μm long, median segment with 1 pair of hair-like setae on dorsal surface, each 10–20 μm long, apical segment with 3 pairs of hair-like setae, 12–17 μm long. **Spiracles** subequal in size; peritreme about 3–5 μm diameter; crescent shaped sclerosis 15–25 μm long. **Bilocular pores**, oval, each 2 μm long and 1 μm wide, with 1 present submarginally about level of each spiracle. **Quinquelocular pores** each 3 μm diameter, as follows; 1 just anterior to each spiracle; 1 medially to each coxa; 2 on each of abdominal segments IV, V, VI. **Microspines** present on median and submedian areas of each abdominal segment, in 2–4 transverse rows; each spine about 1 μm long. **Setae** 1 pair, similar in size and shape to marginal setae, between anterior apex of body and basal segments of antennae; 6 interantennal setae, each 20–30 μm long between basal segments of antennae and anterior apex of clypeus; 1 seta 8–15 μm long medially to each coxa; 2 longitudinal rows, each with 7–8 setae, similar in shape to marginal setae but shorter, each about 8 μm long, extending submarginally from laterad to metathoracic coxae to anterior margin of anal ring, no setae present on most posterior abdominal segment; 2 longitudinal rows of 6 setae, each about 5 μm long, located submarginally on abdomen; 2 longitudinal rows of 4 setae, each about 10 μm long, located submedially on abdomen; plus 2 longitudinal rows of 6 setae, about 17–18 μm long, located medially. **Anal ring** located ventrally; composed of 2 semi-circles; diameter 15–25 μm; each semi-circle with about 17 cells and 3 pointed setae, subequal in size, each 20–30 μm long. Also with a pair of setae, each 12–15 μm long, anterior to anal ring and 1 pair, each 15–25 μm long, postero-laterally to anal ring. **Anal lobes** well-developed; each lobe bearing 2 pointed setae, 12–15 μm long, distinctly thicker than marginal setae, plus 1 pair of long, apical setae 77–125 μm long.

Results

Molecular identification and relationships

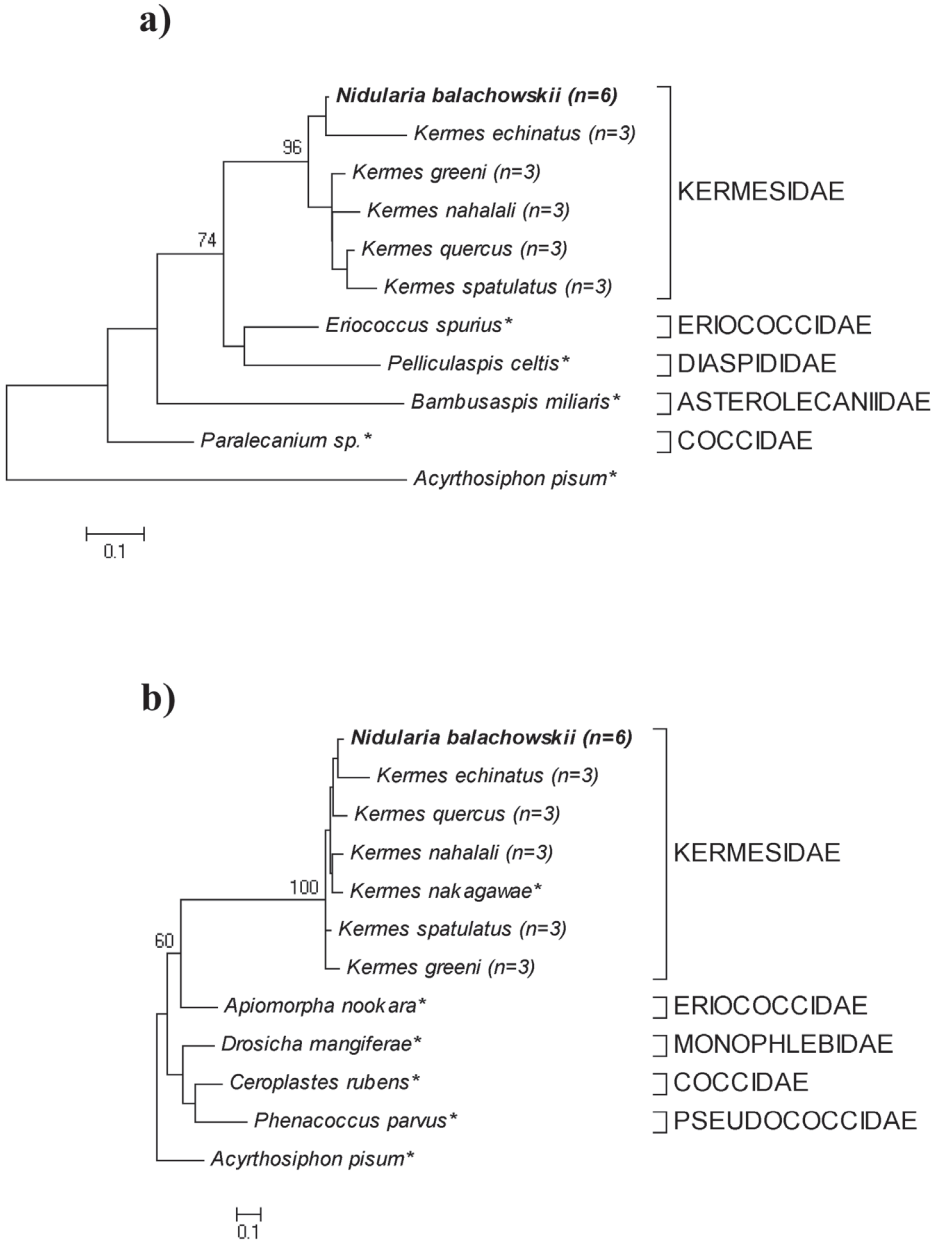

Figure 5. Maximum likelihood trees of 28S (**a**) and COI (**b**) nucleotide sequences of *N. balachowskii* and other Coccoidea species. *Acyrthosiphon pisum* (Aphididae) sequences are used as outgroup species for both trees. Trees were constructed using K2P distance model and numerical values are bootstrap support, based on 1000 replicates (n= number of replicates, * = sequences derived from GenBank).

Molecular results

We obtained a total of forty-two nucleotide sequences from the 28S and COI genes from *N. balachowskii* (six individuals for each gene) and from five adult female Palaearctic Kermesidae species (three individuals for each species for each gene). 28S gene sequences (~700 bp) and COI sequences (~400 bp) from all species were recovered and aligned with sequences of Coccoidea species representing different families (obtained from GenBank). All species for which multiple specimens were sampled showed no interspecies variation. The maximum likelihood analysis of both genes resulted in tree typologies that show that *N. balachowskii* is a distinct species within the monophyletic Kermesidae. *N. balachowskii* is grouped together with other kermesid species and not with the other Coccoidea (Figures 5a+b). The bootstrap value that represents the separation between species of Kermesidae and species from other Coccoidea families is higher in the 28S tree typology, 74, compared to 60 obtained from the COI sequences.

Sequence divergence based on Kimura 2-parameter pairwise distance, between *N. balachowskii* and the other five Kermesidae species ranged from 0.16–0.19 in the 28S gene region. This range is compared to the 0.2–0.3 sequence divergence range between *N. balachowskii* and species from the four other Coccoidea families. In the COI gene region, the sequence divergence between *N. balachowskii* and the six other Kermesidae species ranged from 0.06–0.13 and between *N. balachowskii* and the four species from other Coccoidea families had a sequence divergence range of 0.8–1. Both trees show a strong relationship between *N. balachowskii* and *K. echinatus*, indicating that they are closer to each other than to the other Kermesidae species examined.

Life history

Observations about the life history of *N. balachowskii* were made in three nature reserves in northern Israel: Alonei Abba Nature Reserve, Horshat Tal Nature Reserve and Yehudiya Nature Reserve. The predominant oak species growing in these reserves is *Q. ithaburensis*. In Alonei Abba Reserve, *Q. calliprinos* trees are also present but they are less common. In Israel, *N. balachowskii* has only been found on the trunks and branches of *Q. ithaburensis*, where *N. balachowskii* is an oviparous and univoltine species.

Gravid females were observed on branches and trunks of trees throughout March, during which time they oviposited 200 to 250 (range from 10 specimens) whitish eggs. Each egg was about 0.4 mm long and 0.2 mm wide. Once all of the eggs have been laid and the brood chamber full of eggs, the female dies and the dorsum becomes sclerotized. The sclerotized, convex body of the dead, post-reproductive female may remain on the host tree for a year or more after first-instar emergence.

Eclosion of first-instar nymphs occurs inside the brood chamber and nymphs emerge from the cavity under the dead female body. This takes place from end of March and throughout April. Crawlers settle in bark crevices on branches and on the trunks of the trees. Young teneral females are found on the branches from June to Feb-

ruary. The females continue feeding and increase in body size throughout this period. Feeding was confirmed by observations of honeydew elimination. By late February, the dorsum of the female begins to expand greatly, increasing in convexity and sclerotization. The ventral surface of the abdomen becomes concave, forming the brood chamber into which the eggs are deposited. The ovipositing female secretes a woolly, white wax that surrounds its body margin. No injury has been observed to the oak hosts by *N. balachowskii* in Israel.

Discussion

Life history

We compared our observations of the host plant and development of *N. balachowskii* in Israel to Bodenheimer's 1941 records of this species. In Israel, this scale insect has only been found on the trunks and branches of *Q. ithaburensis* trees, whereas Bodenheimer gives *Quercus* sp. as the host tree in Turkey (Bodenheimer 1941) and Iran (Bodenheimer 1944). The geographical distribution of *Q. ithaburensis* is wide, extending also to Turkey and Iran, and so we may speculate that Bodenheimer's *Quercus* sp. is probably *Q. ithaburensis* ssp. *macrolepis* (Dufour-Dror and Ertas 2004).

We observed that *N. balachowskii* is an oviparous, univoltine species in Israel, similar to Bodenheimer's (1941) observations in Turkey. Earlier observation on the other two species of *Nidularia* indicated that both *N. pulvinata* in Italy (Viggiani 1991) and *N. japonica* in China (Liu et al. 1997) are univoltine. All three species of *Nidularia* have only been recorded so far on oak trees (Ben-Dov et al. 2012). Koteja (1980) redescribed *N. pulvinata* and noted that young specimens were covered with a fragile layer of wax and that, during expansion of the dorsum, this layer breaks into pieces and the females then secrete a nest-like ovisac ventrally and laterally. Kuwana (1918) and Liu et al. (1997) both describe a nest-like ovisac for female *N. japonica*. In Israel, the teneral adult female of *N. balachowskii* also produces a thin layer of dorsal wax but gravid females do not produce a nest-like ovisac. The eggs of *N. balachowskii* are deposited into the egg cavity beneath the venter of the female, as described by Bullington and Kosztarab (1985) and Podsiadlo (2005a) for other kermesid species.

Morphological characters of adult female

Some morphological characters of adult female *N. balachowskii* are compared with those of *N. pulvinata, Kermes quercus* and *K. roboris* (type species of *Kermes*) in Table 2, in order to evaluate the generic and family placement of the former species. All four species possess the following synapomorphic traits: three-segmented labium, bilocular pores on the venter, simple pores on the dorsum, quinquelocular pores surrounding

Table 2. Morphological characters of adult females of *Nidularia balachowskii, N. pulvinata, Kermes quercus* and *K. roboris.*

Character	*Nidularia balachowskii*	*Nidularia pulvinata*	*Kermes quercus*	*Kermes roboris*
antennal segments	1	1	4	5–6
labium segments	3	3	3	3
locular pores on spiracle peritreme	absent	present	absent	absent
locular pores surrounding spiracles	present	present	present	present
legs	absent	absent	present	present
bilocular pores on venter	present	present	present	present
simple pores on dorsum	present	present	present	present
setae -pore clusters on venter	absent	absent	present	present
tubular ducts on venter	1 type	1 type	2 types	2 types
anal ring location	ventral	ventral	dorsal	dorsal
anal ring shape	2 semi- circles	2 semi-circles	circular, complete	2 semi-circles
anal ring cells	present	present	absent	absent
anal ring setae	3 pairs setae	3 pairs setae	setae absent, rare with 2 setae	setae absent, rare with 2 setae

the spiracles, and tubular ducts on the venter. These characters are some of the synapomorphic characters of kermesid adult females that have been described by Ferris (1955), Koteja (1980), Bullington and Kosztarab (1985) and Hodgson (1997).

Within the genus *Nidularia*, adult female *N. balachowskii* share with *N. pulvinata* the following characters: (i) one-segmented antennae; (ii) absence of legs; (iii) absence of setae-pore clusters on venter; (iv) ventral position of anal ring, and (v) an anal ring with setae and cells, whereas the two *Kermes* species have: four, five or six-segmented antennae, and possess legs, setae-pore clusters on venter, anal ring placed on dorsum, and an anal ring without setae and cells. Comparing *N. balachowskii* and *N. pulvinata*, the most obvious distinguishing feature is the presence of quinquelocular pores on the spiracle peritreme of *N. pulvinata.*

Morphological characters of first-instar nymphs

The morphological characters of first-instar nymphs of Nearctic kermesids were summarized by Baer and Kosztarab (1985). Kuwana (1931) and Hu (1986) outlined some distinctive characters for separating first-instar nymphs of Oriental species of kermesids, and Balachowsky (1950, 1953) reviewed the characters of first-instar nymphs of some Palaearctic kermesids.

The first-instar nymphs of *N. balachowskii* share the following characters with other Kermesidae species: (i) six-segmented antennae; (ii) three-segmented labium; (iii) simple pores forming longitudinal lines on the dorsum (iv) dorsal setae; (v) anal

ring with cells and setae; (vi) microspines in rows on abdominal segments, and (vii) bilocular pores on venter. This last character is sometimes overlooked.

Nidularia balachowskii can be distinguished from other Kermesidae species by the form of its marginal setae. *Nidularia balachowskii* has sharply spinose, apically pointed and slightly curved setae, each 9–15 μm long. This differs from *N. pulvinata* which has hair-like setae (Koteja 1980) and *N. japonica* which has setose setae that are somewhat conical at the base (Kuwana 1918, Liu et al. 1997). The first-instars of Palaearctic *Kermes* species that have been described possess conical, hair-like or spatulate marginal setae (Kuwana 1931, Balachowsky 1950, 1953, Sternlicht 1969, Hu 1986, Liu et al. 1997, Podsiadlo 2005b, Pellizzari et al. 2012). There is no unique set of characters that distinguishes the first-instar nymphs of the genus *Nidularia* from the Palaearctic *Kermes* examined.

Key to first-instar nymphs of *Nidularia* species*

1 Quinquelocular pores absent on venter of abdomen...***N. pulvinata* (Planchon)**
– Quinquelocular pores present on venter of abdomen.....**2**
2 Marginal setae sharply spinose, apically pointed and slightly curved; quinquelocular pores, six, on venter of thorax only...***N. balachowskii* Bodenheimer**
– Marginal setae setose and somewhat conical at base; quinquelocular pores, eight, on venter of head and thorax.....***N. japonica* Kuwana**

* Characters of *N. japonica* based on illustrations and descriptions by Kuwana (1918) and Liu et al. (1997).

Molecular analysis

The DNA-sequence data for *N. balachowskii*, six other species of Palaearctic Kermesidae, and species representing six other Coccoidea families showed that gene fragments of both COI and 28S separated *N. balachowskii* from other Coccoidea species, and clearly placed *N. balachowskii* in the Kermesidae. This study confirms that *N. balachowskii* is a distinct species, clearly distinguishable from other closely-related kermesid species.

Conclusion

Based on both the morphological and molecular studies of *N. balachowskii,* the identity of *N. balachowskii* and its placement in the Kermesidae has been substantiated, and we have shown that *N. balachowskii* is congeneric with *N. pulvinata.*

Acknowledgements

This work was funded by Keren Kayemeth LeIsrael (Project # 131-1621-11) and the Israel Taxonomy Initiative. This manuscript is part of the PhD thesis for the senior author. *We thank those who helped us obtain specimens and slide material for this study:* Carl-Axel Gertsson (Murarevägen 13, SE-227 30 Lund, Sweden), Bora Kaydan (Yuzuncu Yil Universty, Agriculture Faculty, Plant Protection Dept. Kampus, Van, Turkey); Jon Martin (BMNH); Imre Foldi and Daniele Matile-Ferrero (MNHN); Elzbieta Podsiadlo (Agricultural University of Warsaw, Department of Zoology, Ul. Ciszewskiego 8, 02-766 Warszawa, Poland). We also thank Adi Faigenboim, Ionit Iberkleid and Adi Kliot for phylogenetic analysis consultation and Alex Protasov for his photographic skills and technical support (Volcani Center, P.O. Box 6, Bet Dagan 50250, Israel). Collection permits for nature reserves were kindly provided by the Israel Nature and Parks Authority.

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ZooKeys 254: 47–65 (2012)
doi: 10.3897/zookeys.254.4223
www.zookeys.org

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A new species of *Amara* (Coleoptera, Carabidae, Zabrini) from Sichuan Province, China, with additional records for other *Amara* species from the region

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Academic editor: *L. Penev* | Received 30 October 2012 | Accepted 11 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:ABE57D06-5BAB-4BF6-A67D-0DFE5895F103

Citation: Hieke F, Kavanaugh DH, Liang HB (2012) A new species of *Amara* (Coleoptera, Carabidae, Zabrini) from Sichuan Province, China, with additional records for other *Amara* species from the region. ZooKeys 254: 47–65. doi: 10.3897/zookeys.254.4223

Abstract

A new species, *Amara* (*Bradytulus) shalulishanica* Hieke & Kavanaugh, **sp. n.** (type locality: Haizishan Yakou, 29.47366°N, 100.21921°E, 4623 m, Shalulishan, Zhuosang Township, Litang County, Sichuan Province, China) is described and diagnosed. Additional records are provided for 16 other *Amara* species, each of which represents one of five different geographical distribution types, which are discussed.

Keywords

Coleoptera, Carabidae, Zabrini, *Amara*, new species, China, Sichuan, Yunnan

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Introduction

Over the ten-year period from 1998 to 2007, two of us (DHK and HBL) had the opportunity to collect extensively in western Yunnan Province, China, as part of a multidisciplinary, international project to inventory the biodiversity of the Gaoligongshan (Gaoligong Mountains). This project also provided the opportunity for us to collect in a few other areas while traveling between Beijing and the study area itself, including most notably one fast traverse in 2007 across several of the mountain ranges that represent the dissected southern edge of the Tibetan Plateau in western Sichuan and northeastern Yunnan Provinces. In total, more than 2,000 specimens representing species of the carabid beetle genus *Amara* Bonelli were collected in the course of this work. Specimens collected in the Gaoligonshan region will form the basis of a separate treatment of the Zabrini of that region (in preparation), which will include a key to the species in that fauna and details of geographical and habitat distributions for each of the included species.

In this contribution, we report on *Amara* species records for specimens collected outside of the Gaoligong region, including those representing one species new to science. That new species is described and a discussion of diagnostic features facilitating discrimination of its adults from those of similar species is provided. One of us (FH) is currently engaged in a comprehenive re-evaluation of subgeneric limits and relationships among *Amara* species, so we refrain from providing a key to subgenera of *Amara* or to species likely related to our new species pending results of that analysis.

Materials and methods

This contribution is based on the study of 279 *Amara* specimens, mainly from Sichuan and Yunnan Provinces. All of these specimens are deposited in the collections of CAS, IZCAS, or ZMHB.

Abbreviations for collections cited in this study follow Evenhuis (2012) as far as possible and include:

BMNH British Museum (Natural History), London, United Kingdom
CAS California Academy of Sciences, San Francisco, U.S.A.
CBAL Collection of A. Baliani, in MCSNG
CCHA Collection of M. de Chaudoir, in MNHN
CFAC Collection of S. Facchini, Torino, Italy
CHEI Collection of W. Heinz, Schwanfeld, Germany
CJED Collection of A. Jedlička, in NMPC
CMEY Collection of P. Meyer, Darmstadt, Germany
CSCI Collection of R. Sciaky, Milan, Italy
CWRA Collection of D. Wrase, Berlin, Germany
DEI Deutsches Entomologisches Institut, Eberswalde, Germany
FRSDD Forest Research Institute, Dehra Dun, India

IZCAS National Zoological Museum of China, Institute of Zoology, Beijing, China
MCSNG Museo Civico di Storia Naturale, Genoa, Italy
MGFT Museum G. Frey, Tutzing, in NHMB
MNHN Muséum National d'Histoire Naturelle, Paris, France
NHMB Naturhistorisches Museum, Basel, Switzerland
NMPC National Museum (Natural History), Prague, Czech Republic
RMNH Rijksmuseum van Natuurlijke Historie, Leiden, Netherlands
SNF Natur-Museum und Forschungs-Institut Senckenberg, Frankfurt am Main, Germany
ZIN Zoological Institute Academy of Sciences, St. Petersburg, Russia
ZMHB Museum für Naturkunde an der Humboldt-Universität, Berlin, Germany
ZSM Zoologische Sammlung des Bayerischen Staates, Munich, Germany

The only measurement recorded is that of body length, taken as a single measure along the midline from the anterior margin of the labrum to the apex of the longer elytron. Information presented on the overall geographical distributions of species is based on the Catalogue of Palaearctic species (Hieke 2003a) and on data about these species gleaned from specimens in the collection at ZMHB.

New species

Amara (Bradytulus) shalulishanica **Hieke & Kavanaugh, sp. n.**
urn:lsid:zoobank.org:act:DEF0D9F5-F136-4561-A0AC-ADE1ABBA9EB7
http://species-id.net/wiki/Amara_shalulishanica
Figs 1–2

Type material. Holotype, a male, in IZCAS, labeled "CHINA, Sichuan, Litang County, Zhuosang Township, Shalulishan, Haizishan Yakou, N29.47366°, E100.21921°"/ "4623 m, 16 September 2007, Stop # 2007-041, D. H. Kavanaugh & H. B. Liang"/ "Holotype *Amara shalulishanica* Hieke and Kavanaugh" [red label]. Paratypes: Total 4 specimens, 1 male and 1 female in CAS, 1 female in IZCAS and 1 male in ZMHB, all with same label data as holotype, except third label reading "Paratype *Amara shalulishanica* Hieke and Kavanaugh" [yellow label]. Type locality: China, Sichuan Province, Litang County, Zhuosang Township, Shalulishan, Haizishan Yakou [29.47366°N, 100.21921°E].

Diagnosis. Adults of *Amara shalulishanica* sp. n. have all the features of other members of subgenus *Bradytulus* Tschitschérine (1894), namely: pronotum with greatest width at or slightly anterior to mid-length; prosternum of male without a punctate fovea at middle; prosternal intercoxal process unmargined and apically asetose; mesofemora bisetose ventrally; mesotibiae of the male with a distinct subapical tooth (seen also in *Curtonotus* males) on medial margin and a brush-like patch of setae ventrally in apical one-fourth. The absence of an apical hook from the right paramere of male genitalia is shared with males of most other *Bradytulus* species.

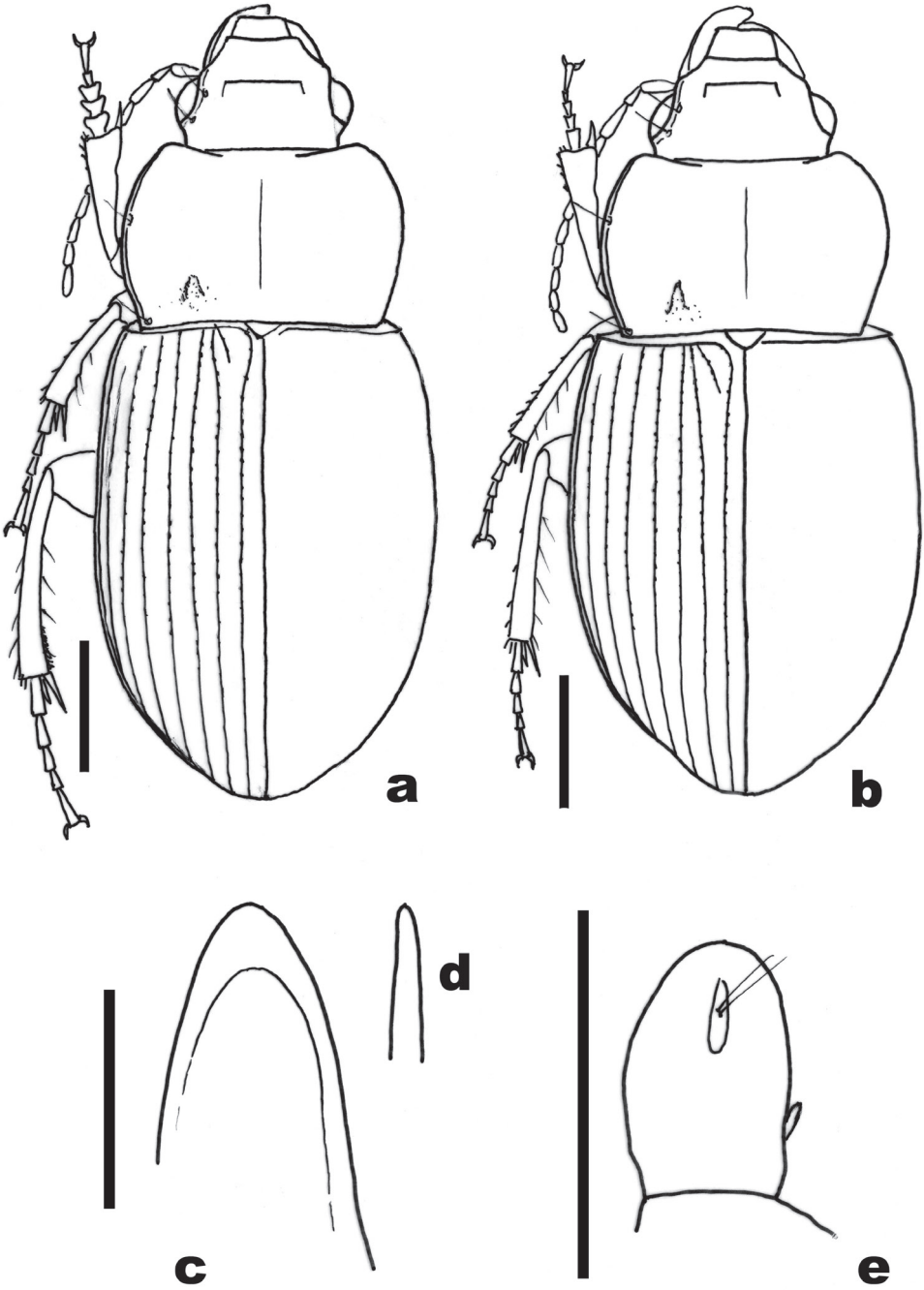

Figure 1. *Amara shalulishanica* sp. n. **a** dorsal habitus of holotype male **b** same of a paratype female **c** apex of the median lobe of aedeagus of holotype, dorsal view **d** same, left lateral view **e** gonostylus of female paratype, ventral view. Scale lines for **a** and **b** = 1.0 mm, for **c–e** = 0.5 mm.

No other species of subgenus *Bradytulus* is known from the Shalulishan (Shaluli Mountains) of Sichuan Province, China. Most species of this subgenus live in the Himalaya Mountains and/or Xizang Province (Tibet) (Hieke 2003b). *Amara thibetana* Tschitschérine, 1894 has been recorded from northern Xizang, Qinghai and Gansu Provinces and may also occur in far northern Sichuan. Its members differ from those of *A. shalulishanica* in being smaller (body length less than 6.0 mm) and having the pronotum with its base more markedly punctate and lateral margins sinuate near the posterior angles. *Amara micans* Tschitschérine, 1894 is widespread in China, especially in Sichuan, and its members differ from those of *A. shalulishanica* in being larger (body length greater than 8.0 mm in most individuals) and having the pronotum with lateral margins sinuate near the posterior angles and the front angles more distinctly extended forward of the anterior margin. Although some brachypterous specimens have been recorded from the Himalayan region, all *A. micans* specimens from Sichuan examined are macropterous and have long metepisterna. The only other *Bradytulus* species with brachypterous members recorded from Sichuan is *Amara platynota* Hieke, 1994 (known from Daxueshan). Its members are larger (body length more than 8.0 mm), have a relatively wider body, broader head and darker legs, and its males have an S-shaped (in dorsal view) median lobe of the aedeagus (Hieke 1994, figs 88–89) and therefore cannot be confused with the new species.

Description. Dorsal habitus as in Fig. 1a-b. Body length male 6.8–7.0 mm, female 6.6–7.0 mm. Color of body dark brown, antennae, palpi and legs reddish brown. Dorsal microscuplture comprised of isodiametric or nearly isodiametric sculpticells throughout, very faintly impressed on head in both sexes, more shallowly impressed on pronotum and elytra in males than in females; males with shinier dorsal luster than females.

Head smooth, broad, with distinct, hemispheric eyes.

Pronotum slightly transverse, with the greatest width slightly anterior to middle and posterior margin narrower than the base of elytra; lateral margins more rounded on anterior half, less arcuate or nearly straight in basal half; posterior margin slightly concave in middle; posterior angles distinct, slightly obtuse, narrowly rounded apically; anterior angles rounded, only slightly extended (about the diameter of the second antennomere) anteriorly beyond the front margin; inner basal foveae formed as short, deeply impressed longitudinal grooves; outer basal foveae absent; basal region with scattered, very fine punctures in and around inner basal foveae. Prosternum of male without a punctate fovea at middle; prosternal intercoxal process smoothly rounded apically, unmargined, asetose apically.

Pterothorax with metepisterna short, not longer than width across anterior margin.

Elytra with slightly curved sides and finely punctate striae; parascutellar striae short, located between striae 1 and 2 and extended from basal margin near base of stria 2 apicomedially toward stria 1; basal borders nearly straight, very slightly arched forward laterally; humeral teeth small but distinct and sharp; umbilicate setal series sparsely and unevenly spaced in the middle region; stria 7 without subapical setiferous pore punctures.

Hind wings short, reduced to a minute scale, hence adults flightless.

Legs with all femora bisetose; mesotibiae of males with a well-developed subapical medial tooth; metatibiae of males with a brush-like patch of setae ventrally in apical one-fourth.

Figure 2. Photographs of type locality for *Amara shalulishanica* sp. n. at Haizishan Yakou [29.47366°N, 100.21921°E], Shalulishan, Zhuosang Township, Litang County, Sichuan Province, China, at an elevation of 4623 m. **a** view looking northeast from the Yakou (Pass) **b** view of area where the type series was collected.

Abdomen with venter only punctate laterally on the sternites 2 and 3. Male with one pair and female with two pairs of anal setiferous pore punctures at the apical margin of the last visible sternite.

Male genitalia with median lobe of aedeagus relatively broad, with apex rounded, apical lamella wider than long in dorsal view (Fig. 1c); right (longer) paramere without apical hook.

Female genitalia with gonostyli broadly oval (Fig. 1d), each with a short basolateral ensiform seta and an subapicoventral nematiform seta.

Etymology. The species epithet, *shalulishanica*, is a Neolatin feminine adjective derived from the mountain range in which the type specimens were collected.

Geographical distribution. Known only from the type locality, where adults were found in a high alpine meadow at 4500 m elevation; probably endemic to the central Shalulishan SSW of Litang.

Habitat distribution. All five specimens of the type series were collected within the area shown in Fig. 2b, under stones on barren substrate interspersed with areas of sparse, low, dry tundra vegetation. Stones under which beetles were found, even in more barren areas, often had accumulations of fine-scale vegetative debris, probably deposited there by wind. Specimens of the new species were collected along with those of two other *Amara* species, *A. micans* Tschitschérine and *A. litangensis* Hieke, 1994, in the same habitat.

Remarks. All but one specimen (the smallest female) of the type series are teneral. Consequently, the color characteristics provided in the description are based solely on that one fully pigmented female specimen. However, because that specimen represents the far end (6.6 mm) of the size range of the type series (all others range between 6.8 and 7.0 mm in length), the holotype male was selected from among those other specimens despite its teneral condition. One consequence of this selection is that detailed structure of the internal sac of the male aedeagus, which typically includes more darkly pigmented or more heavily sclerotized features, cannot be distinguished in the holotype.

Locality records for other *Amara* species

***Amara (Amara) kingdoni* Baliani, 1934**
http://species-id.net/wiki/Amara_kingdoni

Amara (Amara) kingdoni Baliani, 1934b:243. Redescribed by Hieke 2002:647. Type material: Holotype male in BMNH. Type locality: Xizang (Tibet), SE Nagong, Shiuden Gompa, 13000 - 14000 ft.

Amara (Amara) sinica Hieke, 1997:247. Type material: Holotype male in ZMHB and 21 paratypes from various localities in Shaanxi, Sichuan and Qinghai Provinces in CFAC and ZIN. Type locality: China, Sichuan, Wa Shan ("Wanschan"). Synonymized by Hieke 2002:647.

Specimens examined. Total of 14 specimens from the following localities: "CHINA, Yunnan, Shangrila County, Jiantang Township, Shikashan, Napahai Houshan Yakou, 3590 m, N27.93404°/ E099.61287°,"/"17 September 2007, Stop# 2007-042, D.H. Kavanaugh & H.B. Liang collectors", 11 males and 1 female (CAS, IZCAS,

ZMHB); "CHINA, Yunnan, Shangrila County, Xiaozhongdian Township, Tianbaoshan, N27.58517°/ E099.87586°,"/ "3530 m, 18 September 2007, Stop# 2007-043, D.H. Kavanaugh & H.B. Liang collectors", 2 males (CAS, IZCAS).

Geographical distribution. Known only from China (Gansu, Qinghai, Sichuan, Xizang and Yunnan Provinces).

Amara (Pseudoamara) birmana Baliani, 1934

http://species-id.net/wiki/Amara_birmana

Amara (Amara) birmana Baliani, 1934a:189. Type material: Holotype female in BMNH, 1 paratype female in CBAL. Type locality: Burma [without specific locality, but probably from the mountains of northern Myanmar near the border with Yunnan Province, China]. Transferred to subgenus *Pseudoamara* Baliani by Hieke 2002:624.

Amara (Celia) yunnana Baliani, 1934a:193. Type material: Holotype male and 6 paratypes in BMNH. Type locality: China, Yunnan, Yunnan-fou. Synonymized by Hieke 1975:333.

Amara (Pseudoamara) beesoni Baliani, 1934a:190. Type material: Holotype male and allotype in BMNH, 4 paratypes in BMNH and MCSNG. Type locality: India, Assam, Shillong. Synonymized by Hieke 1975:285.

Specimens examined. One male specimen (CAS) from the following locality: "CHINA, Yunnan, Lijiang County, Jiuhe Township, Laojunshan, 3510 m, N26.65079°/ E099.77449°,"/ "21 September 2007, Stop# 2007-048, D.H. Kavanaugh & H.B. Liang collectors".

Geographical distribution. Known from China (Yunnan Province), India (Assam and Sikkim) and Myanmar.

Amara (Celia) rupicola C. Zimmermann, 1832

http://species-id.net/wiki/Amara_rupicola

Amara (Celia) rupicola C. Zimmermann, 1832:26. Type material: Number of syntypes not specified, whereabouts of type series unknown, probably lost; neotype not yet designated. Type locality: Russia, mountains of southern Siberia.

Amara (Celia) marginicollis A. Morawitz, 1863:259. Type material: Number of syntypes not specified, possibly several (4 localities given in original description), probably in ZIN; lectotype not yet designated. Type locality: Russia, Siberia, northern Baikal (locality listed in description). Synonymized by Tschitschérine 1899:164.

Amara ambigena Bates, 1878:716. Type material: Number of syntypes not specified, but probably only a holotype in BMNH (Andrewes 1930:24: "…Type seen…"). Type locality: India, Kashmir, Pangong Valley "Between Tanktze and Chagra" (Andrewes 1930:24). Synonymized by Andrewes 1930:24.

Amara (Celia) tiruka Andrewes, 1924:97. Type material: Lectotype male (designated by Hieke 1983:362) and 2 paralectotypes in BMNH. Type locality: India, Kashmir, Sonemarg and Ladakh, Leh. Synonymy suggested by Hieke 1981:203, formally synonymized by Hieke 1983:362.

Amara (Celia) faucium Andrewes, 1930:24. Type material: 7 syntypes in BMNH; the specimen cited as "Type" should be considered the holotype, specimens cited as "Cotype" should be considered as paratypes. Type locality: China, Xizang (southern Tibet), Jelep La, Phari, Tuna and Kampa Dzong. Synonymized by Hieke 1975:300.

Amara (Celia) rubroangulata Baliani, 1938:186. Type material: Holotype male and allotype in CBAL, 3 female paratypes in BMNH. Type locality: China: Sichuan and Xizang (eastern Tibet): "Lhodzong Poshö", 12600 ft. Synonymized by Hieke 1981:202.

Amara (Amara) prochazkorum Jedlička, 1967:108. Type material: Holotype male in NMPC. Type locality: Mongolia, Karakorum village. Synonymized by Hieke 1999a:181.

Specimens examined. Total of 13 specimens from the following localities: "CHINA, Sichuan, Kangding County, Xinduqiao Township, Liqi He, 3450 m, N30.02448°/ E101.52490°,"/ "14 September 2007, Stop# 2007-035, D.H. Kavanaugh & H.B. Liang collectors", 6 males and 4 females (CAS, IZCAS, ZMHB); "CHINA, Sichuan, Litang County, Gaocheng Township, Daxushan, Highway 318 at Km 3089, 4100 m, N30.03129°/ E100.33336°,"/ "16 September 2007, Stop# 2007-040, D.H. Kavanaugh & H.B. Liang collectors", 1 male and 2 females (CAS, IZCAS).

Geographical distribution. Known from China (Gansu, Qinghai, Shaanxi, Sichuan, Xinjiang and Xizang Provinces), India (Jammu and Kashmir), Kazakhstan, Russia (from the Altai Mountains to Jakutia) and Turkmenistan(?).

***Amara (Xenocelia) mandarina* Baliani, 1932**
http://species-id.net/wiki/Amara_mandarina

Amara (Bradytus) mandarina Baliani, 1932:15. Type material: Holotype male and 3 paratypes in MCSNG, 21 paratypes in BMNH, DEI, NMPC, RMNH, SNF and ZMHB. Type locality: China, Sichuan, Kangding ("Tatsienlu-Chiulung"). Erroneously synonymized with *A. singularis* Tschitschérine by Hieke 1975:317; returned to species status and transferred to subgenus *Xenocelia* by Hieke 2001:104.

Specimens examined. One male specimen (IZCAS) from the following locality: "CHINA, Sichuan, Kangding County, Xinduqiao Township, Gaoersishan, 1 km W of Yakou at Highway 318, 4245 m, N30.04988°/ E101.37485°,"/ "15 September 2007, Stop# 2007-036, D.H. Kavanaugh & H.B. Liang collectors".

Geographical distribution. Known only from China (Sichuan and Yunnan Provinces).

Amara (Pseudocelia) collivaga **Hieke, 1997**
http://species-id.net/wiki/Amara_collivaga

Amara (Bradytus) collivaga Hieke, 1997:200. Type material: Holotype male in ZMHB, 1 paratype female in CHEI. Type locality: China, W. Sichuan, 30 km W of Kangding, 5000 m. Transferred to subgenus *Pseudocelia* by Hieke 2005:155.

Specimens examined. Total of 6 specimens from the following locality: "CHINA, Sichuan, Kangding County, Lucheng Township, Zheduoshan at yakou on Highway 318, 4316 m, N30.07419°/ E101.80413°,"/ "14 September 2007, Stop# 2007-033, D.H. Kavanaugh & H.B. Liang collectors", 2 males and 4 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known only from China (Sichuan Province).

Amara (Bradytus) chalciope **(Bates), 1891**
http://species-id.net/wiki/Amara_chalciope

Leiocnemis chalciope Bates, 1891:71. Type material: Holotype male in BMNH. Type locality: China, Sichuan, Schneeberge bei Kangding ("Snowy Range, near Tatsienlu"), 13000 ft.

Amara (Niphobles) szetschuana Jedlička, 1934a:17. Type material: Holotype female in CJED. Type locality: China, Sichuan, Kangding ("Tatsienlu"). Transferred to subgenus *Bradytus* by Baliani 1937:176. Synonymized by Hieke 1983:361.

Specimens examined. Total of 33 specimens from the following localities: "CHINA, Sichuan, Kangding County, Xinduqiao Township, Gaoersishan, 1 km W of yakou at Highway 318, 4245 m, N30.04988°/ E101.37485°,"/ "15 September 2007, Stop# 2007-036, D.H. Kavanaugh & H.B. Liang collectors", 3 males and 4 females; "CHINA, Sichuan, Luding County, Moxi Township, Hailuogou National Park, NE slope of Gongga Shan, 3035-3220 m, N29.57393°/ E101.99204° to"/ "N29.56721°/ E101.97979°, 12 September 2007, Stop# 2007-031, D.H. Kavanaugh & H.B. Liang collectors", 5 males and 2 females (CAS, IZCAS); "CHINA, Sichuan, Yajiang County, Hekou Township, Daxushan, Jianziwan Yakou at Highway 318, 4400-4450 m, N30.01203°/ E100.85983°, "/ "15 September 2007, Stop# 2007-037, D.H. Kavanaugh & H.B. Liang collectors", 4 males and 5 females (CAS, IZCAS, ZMHB); "CHINA, Sichuan, Yajiang County, vicinity of Daoban work station 135 on road from Litang to Yajiang, N30.1399°/ E100.7068°,"/ "4130 m, under stones in ditch along road adjacent to *Kobresia* meadow, 3 August 2006 B. Bartholomew collector", 2 males and 8 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known from Bhutan and China (Fujian, Sichuan, Xizang and Yunnan Provinces).

***Amara (Bradytus) dissimilis* Tschitschérine, 1894**
http://species-id.net/wiki/Amara_dissimilis

Amara (Bradytus) dissimilis Tschitschérine, 1894:404. Type material: Holotype male in ZIN. Type locality: China, Gansu, Ponggartang ("Thibet sept., Amdo, village Ndàmi"). [Note: The holotype was erroneously labeled "*Brad. dissors* Tschit. 1894 typ!" by Hieke (1999a:165)].

Amara (Bradytus) emmerichi Baliani, 1932:14. Type material: Holotype male ("type") and one paratype in CBAL, additional paratypes in DEI (Döbler 1975:112), NMPC and ZMHB. Type locality: China, Sichuan, Kangding ("Tatsienlu-Chiulung"). Synonymized by Hieke 1999a:165.

Amara (Bradytus) lama Baliani, 1934c:110. Type material: Holotype female and 1 paratype in BMNH, 2 paratypes in CBAL. Type locality: SO.Tibet: Rong Tö Valley, 4000 - 7000 ft. Synonymized by Hieke 1997:225.

Amara (Bradytus) komala Jedlička, 1934b:116. Type material: Holotype female in CJED, 1 paratype female in CMEY. Type locality: China, Yunnan, Longchuan Jiang ("Soling-ho" Valley). Synonymized by Hieke 1995:297.

Amara (Bradytus) mera Jedlička, 1934b:116. Type material: Holotype female and 1 paratype female in CJED. Type locality: China, Yunnan, "Yunnan-fou". Synonymized by Hieke 1995:297.

Specimens examined. Total of 54 specimens from the following localitities: "CHINA, Yunnan, Lijiang County, Jiuhe Township, Laojunshan, 3500 m, N26.64210°/ E099.76745°,"/ "20 September 2007, Stop# 2007-046A , D.H. Kavanaugh & H.B. Liang collectors", 3 males and 3 females, (CAS, IZCAS); "CHINA, Yunnan, Shangrila County, Jiantang Township, Shikashan, Napahai Houshan Yakou, 3590 m, N27.93404°/ E099.61287°,"/"17 September 2007, Stop# 2007-042, D.H. Kavanaugh & H.B. Liang collectors", 27 males and 16 females (CAS, IZCAS, ZMHB); "CHINA, Yunnan, Shangrila County, Xiaozhongdian Township, Tianbaoshan, N27.58517°/ E099.87586°,"/ " 3530 m, 18 September 2007, Stop# 2007-043, D.H. Kavanaugh & H.B. Liang collectors", 1 male and 4 females (CAS, IZCAS).

Geographical distribution. Known only from China (Gansu, Qinghai, Shaanxi, Sichuan, Xizang and Yunnan Provinces).

***Amara (Bradytus) sinuaticollis* A. Morawitz, 1863**
http://species-id.net/wiki/Amara_sinuaticollis

Amara (Bradytus) sinuaticollis A. Morawitz, 1863:257. Type material: Number of syntypes not specified, at least two (male and female mentioned); 1 male and 1 female syntypes in ZIN. Type locality: Russia, "Ussuri".

Specimens examined. Total of 4 specimens from the following localities: "CHINA, Beijing, Huairou County, Shayu Township, Yugou Village, Xiangshui He, 218 m,

N40.41614°/ E116.44386°,"/ "26 October 2002, Stop # DHK-2002-052, D.H. Kavanaugh, P. E. Marek, & H.-B. Liang collectors", 1 male and 1 female (CAS, IZCAS); "CHINA, Yunnan Province, Dali Prefecture, Yongping County, Qutong Township, 1600m, N25.42665°"/ "E99.52924°, 25 June 2000, Stop #2000-005, D. H. Kavanaugh & Liang H.-B. collectors", 1 male and 1 female (CAS, IZCAS).

Geographical distribution. Known from China (Fujian, Gansu, Hebei, Shaanxi, Sichuan and Yunnan Provinces), Japan, Korea and Russia (Khabarovsky Krai and Primorsky Krai).

***Amara (Bradytus) pingshiangi* Jedlička, 1957**
http://species-id.net/wiki/Amara_pingshiangi

Amara (Curtonotus) pingshiangi Jedlička, 1957:24. Type material: Lectotype female in CJED (Hieke 1990:238). Type locality: China: "Süd China: Pingshiang", probably Jiangsu Province. Transferred to subgenus *Bradytus* by Hieke 1990:238.

Specimens examined. One male specimen (CAS) from the following locality: "CHINA, Yunnan, Kunming City, Guandu District, Heilongtan, Kunming Institute of Botany Botanical Garden, 1945 m"/ "N25.14035°/ E102.74107°, 16–17 September 2002, Stop # DHK-2002-019, D.H. Kavanaugh & P.E. Marek collectors" [first record for Yunnan Province].

Geographical distribution. Known only from China (Zhejiang, Fujian, Jiangsu and Sichuan Provinces).

***Amara (Bradytulus) micans* Tschitschérine, 1894**
http://species-id.net/wiki/Amara_micans

Amara (Bradytus) micans Tschitschérine, 1894:402. Type material: Lectotype (designated by Hieke 1973:111) and several paralectotypes in ZIN. Type locality: China, northern Qinghai Province, "Amdo-Plateau", probably in the area around the confluence of the Blue and Yellow Rivers (west of Bayanharshan). Transferred to subgenus *Niphobles* by Hieke 1975:308 and to subgenus *Bradytulus* by Hieke 2003b:157.

Amara (Niphobles) splendens Andrewes, 1926:75. Type material: Holotype ("Type") and paratype ("Cotype") in BMNH, another paratype in FRSDD. Type locality: northern India, Himachal Pradesh (Kumaon District), Milam and Burphu in Gori Valley, 11500 ft. Synonymized by Hieke 1981:225.

Amara (Bradytus) pernix Csiki, 1929:457, replacement name for *A. micans* Tschitschérine, 1894 (*nec* Letzner, 1852) [unnecessary change]. Synonymized by Hieke 1981:225.

Amara (Niphobles) jedlickai Baliani, 1932:158. Type material: 5 syntypes in MCSNG and MGFT (lectotype not yet designated). Type locality: China, Sichuan, Kangding ("Tatsienlu-Chiulung"). Synonymized by Hieke 1975:308.

Amara (Balianiia) vafra Lutshnik, 1935:267. Type material: Holotype male ("Monotypus") in ZIN. Type locality: southern Mongolia, Gobi-Altai ("N.-Seite des Chines. Altai"). Synonymized by Hieke 1973:111.

Amara (Bradytus) eberti Jedlička, 1965:101. Type material: Holotype ("Type") and 4 paratypes ("Cotype") in ZSM, 2 additional paratypes in BMNH and 1 in ZMHB. Type locality: Nepal, Khumbu, Khumdzung. Synonymized by Hieke 1975: 298.

Amara (Bradytus) hellmichi Jedlička, 1965:102. Type material: 2 syntypes in ZSM and 1 in NMPC (lectotype not yet designated). Type locality: Nepal, Khumbu, Khumdzung. Synonymized by Hieke 1975:305.

Specimens examined. Total of 11 specimens from the following localities: "CHINA, Sichuan, Litang County, Zhuosang Township, Shalulishan, Haizishan Yakou, N29.47366°/ E100.21921°,"/ "4623 m, 16 September 2007, Stop# 2007-041, D.H. Kavanaugh & H.B. Liang collectors", 7 males and 3 females (CAS, IZCAS, ZMHB); "CHINA, Sichuan, Yajiang County, Hekou Township, Daxushan, Jianziwan Yakou at Highway 318, 4400-4450 m, N30.01203°/ E100.85983°,"/ "15 September 2007, Stop# 2007-037, D.H. Kavanaugh & H.B. Liang collectors", 1 female (CAS).

Geographical distribution. Known from China (Gansu, Qinghai, Sichuan, Xizang and Yunnan Provinces), India (Jammu and Kashmir, Uttar Pradesh), Nepal and Pakistan.

***Amara (Reductocelia) daxueshanensis* Hieke, 2000**
http://species-id.net/wiki/Amara_daxueshanensis

Amara (Reductocelia) daxueshanensis Hieke, 2000:118. Type material: Holotype male and 13 paratypes in ZMHB. Type locality: China, northern Yunnan, Zhongdian area, 3200–3300 m.

Specimens examined. Total of 47 specimens from the following localities: "CHINA, Yunnan, Lijiang County, Jiuhe Township, Laojunshan, 3500 m, N26.64210°/ E099.76745°,"/ "20 September 2007, Stop# 2007-046A , D.H. Kavanaugh & H.B. Liang collectors", 1 female (CAS); "CHINA, Yunnan, Shangrila County, Jiantang Township, Shikashan, Napahai Houshan Yakou, 3590 m, N27.93404°/ E099.61287°,"/"17 September 2007, Stop# 2007-042, D.H. Kavanaugh & H.B. Liang collectors", 3 males and 2 females (CAS, IZCAS); "CHINA, Yunnan, Shangrila County, Xiaozhongdian Township, Tianbaoshan, N27.58517°/ E099.87586°,"/ "3530 m, 18 September 2007, Stop# 2007-043, D.H. Kavanaugh & H.B. Liang collectors", 19 males and 22 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known only from China (Yunnan Province).

***Amara (Reductocelia) dequensis* Hieke, 1999**
http://species-id.net/wiki/Amara_dequensis

Amara (Reductocelia) dequensis Hieke, 1999b:347. Type material: Holotype male and 13 paratypes in ZMHB. Type locality: China, northern Yunnan, Zhongdian area, 3200–3300 m.

Specimens examined. One male specimen (IZCAS) from the following locality: "CHINA, Sichuan, Batang County, pass between Ciwu and Zhong Xinrong on road from Derong to Batang, N30.1399°/ E100.7068°,"/ "4130 m, under stones in oak scrub slope adjacent to *Kobresia* meadow, 29 July 2006 B. Bartholomew collector".

Geographical distribution. Known only from China (Sichuan and Yunnan Provinces).

***Amara (Reductocelia) litangensis* Hieke, 1994**
http://species-id.net/wiki/Amara_litangensis

Amara (Bradytulus) litangensis Hieke, 1994:315. Type material: Holotype male and 8 paratypes in CSCI, 5 paratypes in ZMHB and 2 paratypes in CWRA. Type locality: China, Sichuan, 10 km S of Litang, 4000 m. Transferred to subgenus *Reductocelia* by Hieke 1999b:348.

Specimens examined. Total of 18 specimens from the following locality: "CHINA, Sichuan, Litang County, Disan Township, Shalulishan, Haizishan Yakou, N29.47366°/ E100.21921°,"/ "4623 m, 16 September 2007, Stop# 2007-041, D.H. Kavanaugh & H.B. Liang collectors", 5 males and 13 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known only from China (Sichuan Province).

***Amara (Reductocelia) stricticeps* Baliani, 1932**
http://species-id.net/wiki/Amara_stricticeps

Amara (Celia) stricticeps Baliani, 1932:7. Type material: Holotype male and allotype in CBAL, 1 paratype in DEI (Döbler 1975:142) and also in SNF. Type locality: China, Sichuan, Kangding ("Tatsienlu-Chiulung"). Transferred to subgenus *Leiromorpha* by Baliani 1937:176 and to subgenus *Reductocelia* by Hieke 1999b:356.

Specimens examined. Total of 58 specimens from the following localities: "CHINA, Sichuan, Kangding County, Lucheng Township, Zheduoshan at yakou on Highway 318, 4316 m, N30.07419°/ E101.80413°,"/ "14 September 2007, Stop# 2007-033, D.H. Kavanaugh & H.B. Liang collectors", 1 male (CAS); "CHINA, Sichuan, Kangding County, Xinduqiao Township, Gaoersishan, 1 km W of yakou at Highway 318, 4245 m, N30.04988°/ E101.37485°,"/ "15 September 2007, Stop# 2007-036,

D.H. Kavanaugh & H.B. Liang collectors", 23 males and 16 females (CAS, IZCAS, ZMHB); "CHINA, Sichuan, Litang County, Gaocheng Township, Daxushan, Highway 318 at Km 3089, 4100 m, N30.03129°/ E100.33336°,"/ "16 September 2007, Stop# 2007-040, D.H. Kavanaugh & H.B. Liang collectors", 3 males and 1 female (CAS, IZCAS); "CHINA, Sichuan, Yajiang County, Hekou Township, Daxushan, Jianziwan Yakou at Highway 318, 4400-4450 m, N30.01203°/ E100.85983°,"/ "15 September 2007, Stop# 2007-037, D.H. Kavanaugh & H.B. Liang collectors", 10 males and 3 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known only from China (Sichuan Province).

***Amara (Curtonotus) kangdingensis* Hieke, 1997**
http://species-id.net/wiki/Amara_kangdingensis

Amara (Curtonotus) kangdingensis Hieke, 1997:222. Type material: Holotype male and 1 paratype male in CHEI**,** 1 paratype male in ZMHB. Type locality: China, Sichuan, pass 30 km W of Kangding, 4000–4200 m.

Specimens examined. Total of 13 specimens from the following localities: "CHINA, Sichuan, Kangding County, Lucheng Township, Zheduoshan at yakou on Highway 318, 4316 m, N30.07419°/ E101.80413°,"/ "14 September 2007, Stop# 2007-033, D.H. Kavanaugh & H.B. Liang collectors", 2 males and 3 females (CAS, IZCAS, ZMHB).

Geographical distribution. Known only from China (Sichuan Province).

***Amara (Curtonotus) macronota* Solsky, 1875**
http://species-id.net/wiki/Amara_macronota

Curtonotus nitens Putzeys, 1866:234 (*nec* Sturm, 1825). Type material: Holotype, female, in CCHA. Type locality: northern China [without specific locality ("Chine boréale")].

Curtonotus macronotus Solsky, 1875:265. Type material: Holotype, in ZIN (Tschitschérine 1894:386). Type locality: Russia, Primorsky Krai, Suyfun River at «Nikolskoje». Synonymized by Tschitschérine 1894:385.

Amara (Curtonotus) jureceki Jedlička, 1957:29. Type material: Number of syntypes not specified, but probably only the holotype (based on text of description), in CJED.- Type locality: Russia, Primorsky Krai, Vladivostok. Synonymized by Lafer 1989:180.

Amara (Curtonotus) ovalipennis Jedlička, 1957:30. Type material: Holotype, male, in CJED. Type locality: Japan, Kyoto. Regarded as a subspecies, *A. macronota ovalipennis* Jedlička, by Morita 1987:70. Synonymized by Hieke 1995:322.

Specimens examined. Two female specimens (CAS, IZCAS) from the following locality: "CHINA, Beijing, Wuling Mountains, Miyun County, Xinchangzi Township, Xiakou

Village, small branch of Andamu He, 415 m,"/ "N40.65278°/E117.34069°, 27 October 2002 Stop # DHK-2002-056, D.H. Kavanaugh, P. E. Marek, & H.-B. Liang collectors".

Geographical distribution. Known from China (Gansu, Shaanxi, Sichuan and Yunnan Provinces), Japan, Korea and Russia (Primorsky Krai and Khabarovsky Krai).

Discussion

The diverse carabid beetle fauna of China and adjacent areas remains relatively poorly sampled and many new species are discovered in and described from the region each year. The geographical distributions of virtually all species in the fauna are still poorly known, and this is certainly true for the Chinese *Amara* species. Despite this incomplete knowledge of the fauna, however, the *Amara* species recorded from Yunnan and Sichuan Provinces can be grouped, at least tentatively, into five different distributional types. The species on which we have reported here are listed below according to their apparent distributional type.

1. *Widespread eastern and central Palaearctic species.* These include species occurring in Russia (from the Altai to the Amur region, from Yakutia to the Mongolian border), Mongolia, and northern China (rarely as far south as the Himalaya): *Amara rupicola*.

2. *Macropterous eastern Palaearctic species*. These include species occurring in central Japan, the Ussuri region of the Russian Far East, Korea, eastern Mongolia, and China (excluding the west but often including Taiwan): *Amara sinuaticollis* and *A. macronota*.

3. *Macropterous species widespread in China*. These include species occurring in several Chinese provinces, and in many cases also in regions of the neighboring Himalayan countries of India, Nepal, Bhutan, and Myanmar: *Amara kingdoni*, *A. mandarina*, *A. chalciope*, *A. dissimilis*, *A. pingshiangi*, and *A. micans*.

4. *Macropterous Himalayan species*. These include species occurring at least in the central and eastern parts of this region, and often also in Yunnan, less commonly also in southern Sichuan: *Amara birmana*.

5. *Apterous endemic Chinese species.* These include species occurring only in small areas, mainly in isolated mountain ranges that occupy only part of one province or border areas linking adjacent parts of two provinces: *Amara collivaga*, *Amara shalulishanica* sp. n., *Amara daxueshanensis*, *A. dequensis*, *A. litangensis*, *A. stricticeps*, and *A. kangdingensis*.

As additional sampling throughout the region continues, particularly in remote areas not yet explored, the ranges of known and additional, still undiscovered species will become better known. The appropriateness of recognizing these different distributional types for characterizing the geographical ranges of different *Amara* species, as well as other species of the regional fauna, will be tested by these future findings.

Acknowledgements

Fieldwork for this project was supported by grants from the National Science Foundation of China (Grant No. 30570213 and 31172047) to the Institute of Zoology (Beijing), and from the U. S. National Science Foundation (Grant No. DEB–0103795) to the California Academy of Sciences.

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ZooKeys 254: 67–87 (2012)
doi: 10.3897/zookeys.254.4043
www.zookeys.org

RESEARCH ARTICLE

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Two new genera and species of the termite symbiont lineage Termitohospitini (Coleoptera, Staphylinidae, Aleocharinae) from Bolivia and peninsular Malaysia

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Academic editor: *J. Klimaszewski* | Received 26 September 2012 | Accepted 5 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:0A80E5CF-3E19-4CE5-BD5F-548F659AEF91

Citation: Kanao T, Eldredge KT, Maruyama M (2012) Two new genera and species of the termite symbiont lineage Termitohospitini (Coleoptera, Staphylinidae, Aleocharinae) from Bolivia and peninsular Malaysia. ZooKeys 254: 67–87. doi: 10.3897/zookeys.254.4043

Abstract

Coptotermocola clavicornis **gen. & sp. n.** and *Neotermitosocius bolivianus* **gen. & sp. n.** of the termite inquilinous tribe Termitohospitini are described from peninsular Malaysia and Bolivia, respectively. The Termitohospitini are most readily diagnosable by the distally migrated anterior tentorial pits that are no longer associated with the antennal fossae, and by the enlarged vertex which obscures the antennal fossae dorsally. Additionally, the Termitohospitini are hypothesized to share a recent common ancestor with the Masuriini and Myllaenini due to shared derived morphologies of the lacinia distal teeth with lateral cuticular processes, presence of a unique maxillary palpomere III sensilla, and anterolateral angles of mentum produced. Habitus photographs and illustrations of diagnostic features are provided for the two new genera in order to facilitate future work.

Keywords

Coptotermes, Hetairotermitina, Termitohospitina, limuloid, Masuriini, Myllaenini, physogastry, termitophily

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Introduction

Eusocial insect colonies are often accompanied by a unique faunal assembly that takes advantage of available colony resources (e.g. Hölldobler and Wilson 1990, Wheeler 1928). Symbionts of termites are commonly referred to as termitophiles (Kistner 1969). A large number of termitophiles have evolved within the beetle family Staphylinidae, particularly in lineages of the subfamily Aleocharinae. Among the recognized 58 tribes (Bouchard et al. 2011), 1,151 genera and 12,851 species (Thayer 2005) of Aleocharinae, approximately 650 species and 190 genera among 17 tribes have recorded associations with termites, of which eleven tribes are comprised exclusively of termitophilous species (Kanao unpublished data).

Termitophilous Aleocharinae that exhibit relatively complex symbiosis with their host termites have repeatedly converged on physogastric and limuloid body plans (e.g. Figs 24–27). Physogastry is the result of abdominal or thoracic inflation and is hypothesized to serve a mimetic function (Kistner 1969). Physogastric species are generally well integrated into the termite social system, actively interacting with their hosts and slow moving. On the other hand, limuloid taxa are teardrop-shaped and the body plan is hypothesized to serve a defensive function (Kistner 1979). Limuloid taxa often also exhibit extremely compact appendages and cavities to receive structures more vulnerable to host aggression (e.g. distal podomeres). In contrast to physogastric species, limuloid species are faster moving and usually do not actively interact with their hosts.

Among the aleocharine termitophiles, the tribe Termitohospitini Seevers, 1941 is comprised exclusively of termite symbiotic species. This tribe was originally proposed by Seevers (1941) to separate a handful of Neotropical termitophilous species of the tribe Bolitocharini (presently Homalotini Heer, 1839). Seevers (1957) later expanded Termitohospitini to include termitophiles from other biogeographic regions based on the 4-4-5 tarsal formula and proposed four subtribes: Termitohospitina Seevers, 1941 for Neotropical members; Hetairotermitina Seevers, 1957 for Australian and Oriental members; Termitospectrina Seevers, 1957 for Pantropical members; Termitusina Seevers, 1957 for Ethiopian members. Kistner and Jacobson (1976) subsequently raised Termitusina to tribe with Termitospectrina as a subtribe within it, based on 4-articled maxillary and 3-articled labial palpi. Therefore, Termitohospitini is presently divided into two subtribes, Termitohospitina and Hetairotermitina. Termitohospitina includes 15 species distributed among five genera (Mann 1923, Silvestri 1947, Seevers 1957, Borgmeier 1959) and Hetairotermitina includes 21 species distributed among seven genera (Lea 1910, Cameron 1920, 1950, Seevers 1957, Kistner 1970, 1976, 1985, Abdel-Galil and Kistner 1987, Roisin and Pasteels 1990, Oke 1993, Pace 1993, Maruyama and Iwata 2002).

Most species of Termitohospitini ancestrally share a limuloid body form but a few taxa have secondarily evolved physogastric forms (Kanao et al. in preparation). Notably, the species of *Coptoxenus* Kistner, 1976 exhibits physogastry of the thorax, a peculiar condition among aleocharines. With a diversity of body forms, the tribe offers a unique opportunity to study the independent evolution of adaptive morphologies

as it correlates with integration and behavior with host termite societies. However, as with most termitophiles, basic understanding of Termitohospitini biodiversity lags behind that of their free-living relatives. Inquilinous groups are difficult to collect and relatively few specimens are available for comprehensive understanding of their biodiversity. Here we describe two new genera and species of Termitohospitini as part of our ongoing research on the evolution of termitophily in termitohospitines. We provide habitus photographs, illustrations of diagnostic features, a new tribal diagnosis as an update to previous diagnoses (Seevers 1941, 1957), and putative synapomorphies that support a close relationship of Termitohospitini with Masuriini and Myllaenini.

Materials and methods

The first author conducted fieldwork at Ulu Gombak, which is approximately 25 km northwest of Kuala Lumpur, Malaysia in 2010, 2011 and 2012. Specimens of a new genus collected at the site for this study were initially preserved in 2.0 ml vials of 80% ethanol. The other undescribed genus was discovered in the Snow Entomological Museum Collection at the University of Kansas.

The technical procedures used for this study generally follow those described in Maruyama (2006). When a permanent mount was made, cleared specimens or parts were dehydrated by successively transferring specimens to higher ethanol (EtOH) concentrations until finally reaching 100% EtOH, and then placed into Euparal. When appropriate, dissections were made within Euparal on a glass mounting substrate. Full body dissections were made and preserved on glass microscope slides. Dissected body parts were preserved and mounted on halved glass cover slips, subsequently glued onto a halved paper glue board, and mounted under the respective specimen (see Maruyama 2004, for details).

In the descriptions, macrochaetotaxy formulas depict raw numbers of setae and not the numbers of setal pairs. Additionally, the number of macrosetae on tergite IX and X refers to one side of the body and therefore the numbers of setal pairs.

Table 1. Checklist of described species of Termitohospitini. Asterisks indicate type species.

Tribe Termitohospitini	*borealis* Kistner, 1970
Subtribe Hetairotermitina	*bryanti* Cameron, 1950
***Coptophysa* Roisin & Pasteels, 1990**	*capitalis* Kistner, 1970
obesa Roisin & Pasteels, 1990	*gayi* Kistner, 1970
***Coptophysella* Roisin & Pasteels, 1990**	*greavesi* Kistner, 1970
pulposa Roisin & Pasteels, 1990	*barretti* Cameron, 1950
***Coptotermocola* gen. n.**	*hirsutus* Kistner, 1970
clavicornis **sp. n.**	*insulanus* Seevers, 1957
***Coptoxenus* Kistner, 1976**	*latevricola* (Lea, 1910) *
thronileyi Kistner, 1976	*occidentalis* Kistner, 1970
***Hetairotermes* Cameron, 1920**	*piceus* Cameron, 1920
agilis Cameron, 1920	*punctiventris* (Lea, 1910)

***Japanophilus* Maruyama & Iwata, 2002**
hojoi Maruyama & Iwata, 2002
***Sinophilus* Kistner, 1985**
rougemonti Pace, 1993
xiai Kistner, 1985 *
yukoae Maruyama & Iwata, 2002
***Termitobra* Seevers, 1957**
perinthoides Seevers, 1957
Subtribe Termitohospitina
***Blapticoxenus* Mann, 1923**
brunneus Mann, 1923
***Noetermitosocius* gen. n.**
bolivianus **sp. n.**
***Paratermitosocius* Seevers, 1941**
vestitus (Mann, 1923)

***Termitohospes* Seevers, 1941**
brasiliana Seevers, 1957
guianae Seevers, 1941
limulus Seevers, 1957
miricroniger Seevers, 1941 *
nitens Borgmeier, 1959
panamensis Seevers, 1941
punctulatus Borgmeier, 1959
silvestrii Seevers, 1957
tachyporoides Seevers, 1957
unicolor (Silvestri, 1947)
***Termitosodails* Seevers, 1941**
barticae Seevers, 1941 *
fasciatus (Silvestri, 1947)
***Termitosocius* Seevers, 1941**
microps Seevers, 1941

Taxonomy

Termitohospitini Seevers, 1941

Type genus: *Termitohospes* Seevers, 1941.

Diagnosis. Members of the tribe Termitohospitini most closely resemble the tribe Myllaenini and Masuriini (*sensu* Ahn and Ashe 2004), and are hypothesized to be closely related based on the following combination of putative synapomorphies: 1) subapical marginal setae of maxillary lacinia with basal, paired cuticular processes (spinose scales of Ahn and Ashe 2004); 2) presence of a unique sensory patch on lateral surface of maxillary palpomere III; 3) anterolateral angles of mentum produced.

Species of Termitohospitini are diagnosable from all other Aleocharinae, including Myllaenini and Masuriini, by the following putative synapomorphies: 1) anterior tentorial arms of the head migrated anteriorly and disassociated from antennal fossae (Seevers 1941); 2) antennal fossae dorsally obscured by enlarged vertex (Seevers 1941); 3) maxillary lacinia with basal paired cuticular processes reduced in size; 4) processes of anterolateral angles of mentum reduced in size; 5) ligula broad and reduced in length.

Neotermitosocius Kanao, Eldredge & Maruyama, gen. n.

urn:lsid:zoobank.org:act:7CEBD279-F24B-4BA5-A2F6-2496848C786E
http://species-id.net/wiki/Neotermitosocius

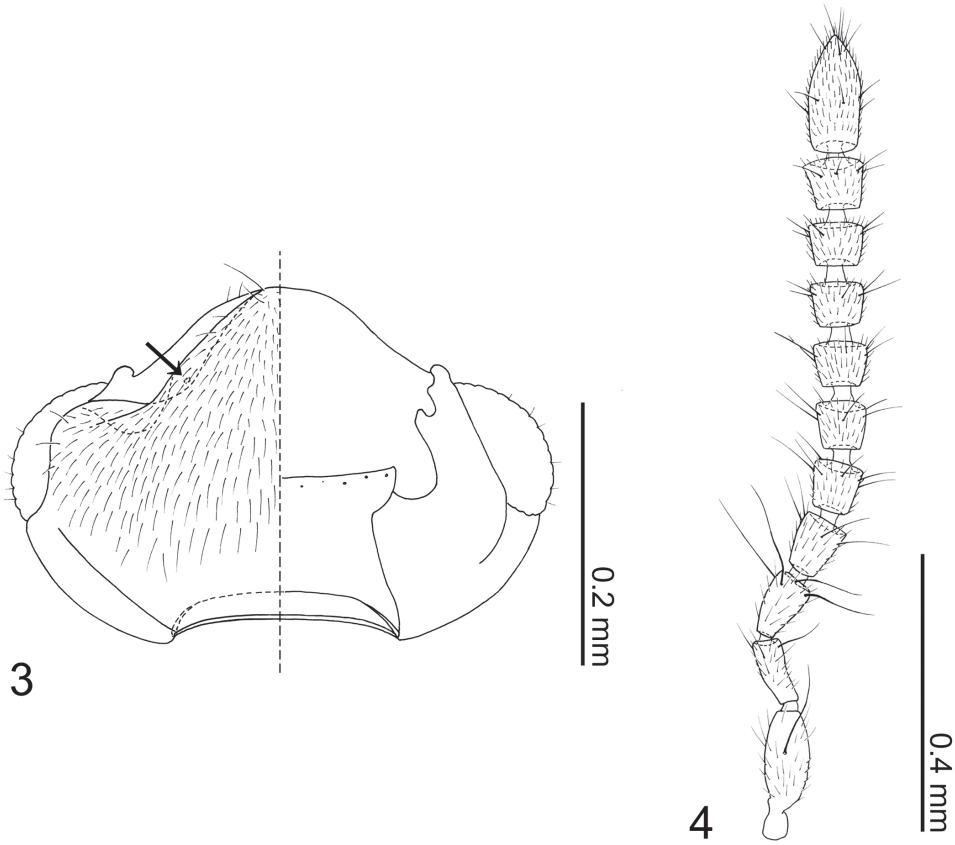
Type species: *Neotermitosocius bolivianus* sp. n.

Diagnosis. This monotypic genus is distinguishable from other Termitohospitini by its slender parallel-sided body, longer legs and apically truncate pronotum, which dorsally exposes much of the head (Fig. 1).

Figures 1–2. Habitus of *Neotermitosocius bolivianus*. **1** dorsal view **2** ventral view.

Description. Overall shape (Fig. 1) parallel-sided and somewhat dorsoventrally flattened. Posterior one-fifth of head covered by pronotum (Fig. 1).

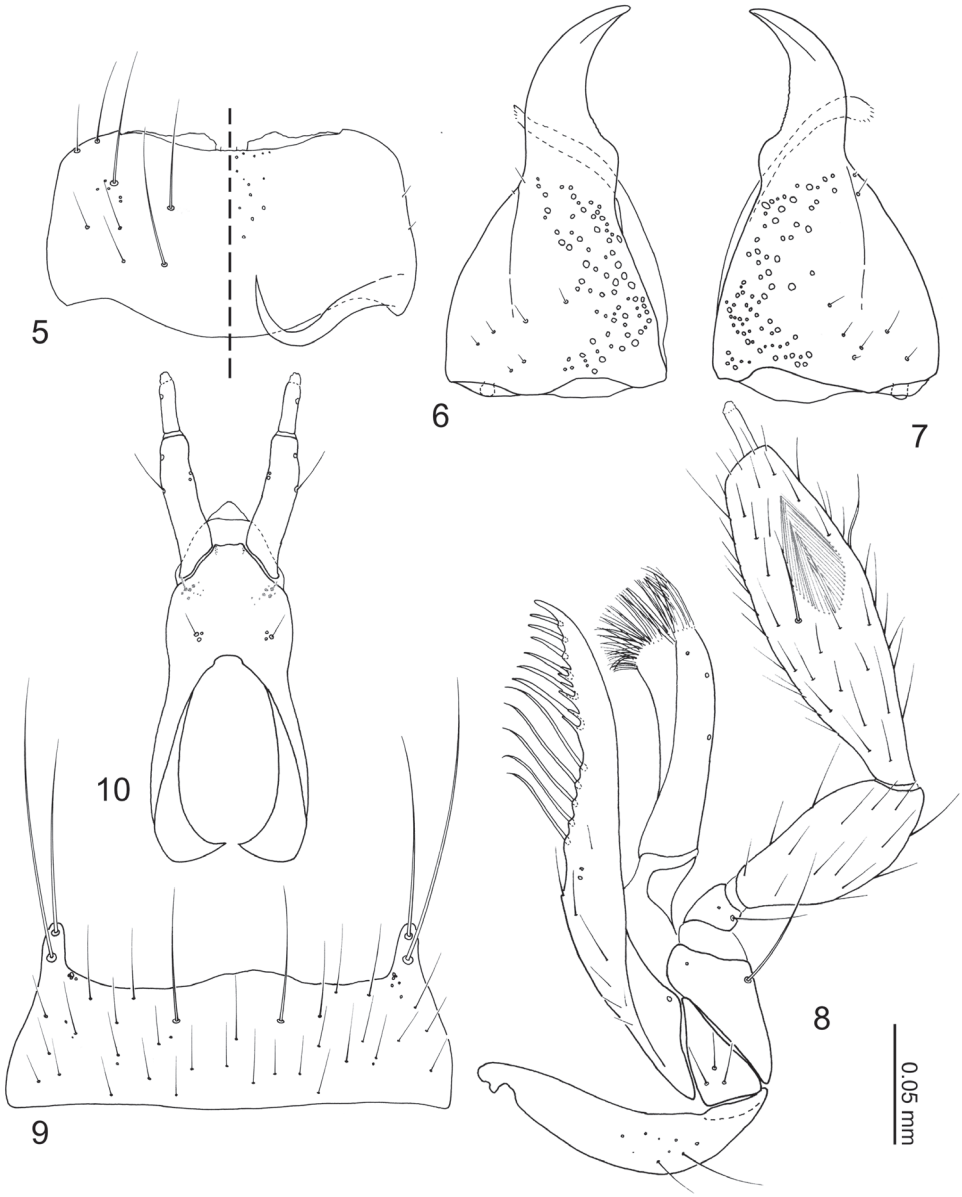
Head (Fig. 3) transverse, widest at eyes. Antennal fossae deep, partially dorsally obscured by vertex. Eyes large and produced anterolaterally. Antennae (Fig. 4) with 11 articles; antennal articles compact and article pedicles scarcely visible externally; antennomeres I and XI longest; antennomere I partially obscured by enlarged antennal fossae; antennomere II approximately three times longer than wide, dilated at apex, narrower than other articles; antennomere III trapezoidal, apex twice as wide as base; antennomere IV trapezoidal, slightly shorter than antennomere III; antennomere V–X successively widening distally; antennomere XI narrowed apically. Labrum (Fig. 5) transverse, with pores at middle, anterior margin of membranous area concave; epipharynx (Fig. 5) relatively smooth, midline with pores from apex to middle. Mandibles (Figs 6–7) almost symmetrical, apex acute, dorsally covered with numerous pores, with anterior one-fifth curved adorally, moderately developed tooth present at middle of adoral margin, base of scrobe strongly laterally developed. Right mandible (Fig. 7) apex slightly more acute. Maxillary (Fig. 8) lacinia elongate, adorally with 11 strong setae; apical three setae shortest, preceding basal three setae with cuticular processes (spinose scale of Ahn and Ashe 2004) on either side of base; galea as long as lacinia, slightly dilated apically, apex densely setulate; palpus with 4 articles; article I trapezoidal; article II oval, narrowed at base; article III approximately twice as long as article II, more than three times longer than wide, widest at middle, sparsely covered with long setae, sensory



Figures 3–4. *Neotermitosocius bolivianus*. **3** head capsule, left side = dorsal view, right side = ventral view **4** antenna.

patch near middle of lateral surface; article IV narrow and parallel-sided, approximately one-fourth as wide as apical margin of article III; pseudosegment poorly delimited and inconspicuous. Mentum (Fig. 9) trapezoidal, sparsely covered with setae and small pseudopores; anterior margin bisinuate, with a pair of long setae at middle; anterolateral corners produced and with 2 long setae; posterior margin almost straight. Labial palpus (Fig. 10) with 3 articles, first article a fusion of I + II; article I + II with a seta (probably homologous with seta h of Sawada [1972]) at outer medial margin, twin and medial pores present; article III half as long as article I, with a pore at anterolateral corner; ligula triangular with posterior half sclerotized; prementum wider than long, disc with a pair of setal pores and two pair of real pores present; apodemes broad and longer than disc, dilated posteriorly with apices recurved internally and almost touching.

Pronotum (Fig. 11) transverse, widest near middle; anterior margin broadly concave; posterior margin slightly rounded. Prosternum reduced in length. Elytra (Fig. 12) subquadrate, longer than wide with lateral margin deflexed. Wings fully developed. Mesoventrite (Fig. 13) short, approximately half as long as metaventrite; mesoventral



Figures 5–10. *Neotermitosocius bolivianus* mouthparts. **5** labrum, left side = labrum, right side = epipharynx **6** left mandible, dorsal view **7** right mandible, dorsal view **8** maxilla, ventral view **9** mentum, ventral view **10** labium in ventral view.

process developed posteriorly and carinate; mesocoxal cavity narrowly separated and marginal bead complete. Legs (Figs 14–16) overall slender; tarsal formula 4-4-5; tarsomeres slender and parallel-sided. Fore leg (Fig. 14) with coxa subequal in length to femur; trochanter subtriangular; femur narrowed apically; tibia thin; tarsomeres I–III

subequal in length, tarsomere IV longer. Mid leg (Fig. 15) with coxa oval; trochanter small; femur narrowed apically; tibia thin, slightly dilated subapically, one macroseta present on dorsal surface; tarsomeres subequal in length. Hind leg with coxa (Fig. 16) subtriangular; trochanter subtriangular, macroseta present on anterior surface; femur slightly narrowed apically; tibia thin, slightly dilated apically; tarsomere I longer than II + III combined, tarsomere V twice as long as IV.

Abdomen (Figs 1–2) narrowed posteriorly. Segment I represented only by tergite I fused to metanotum. Segment II represented only by tergite II. Segments III–VII with 1 tergite, 1 sternite, and 2 pairs of paratergites respectively. Tergite VIII (Fig. 17) narrowed posteriorly, posterior margin slightly medially produced. Sternite VIII (Fig. 18) with posterior margin rounded. Tergite IX (Fig. 19) fully subdivided dorsally by tergite X; tergite X (Fig. 19) fully divided medially to base. Tergite X divided, only connected each other and tergite IX on anterior margin.

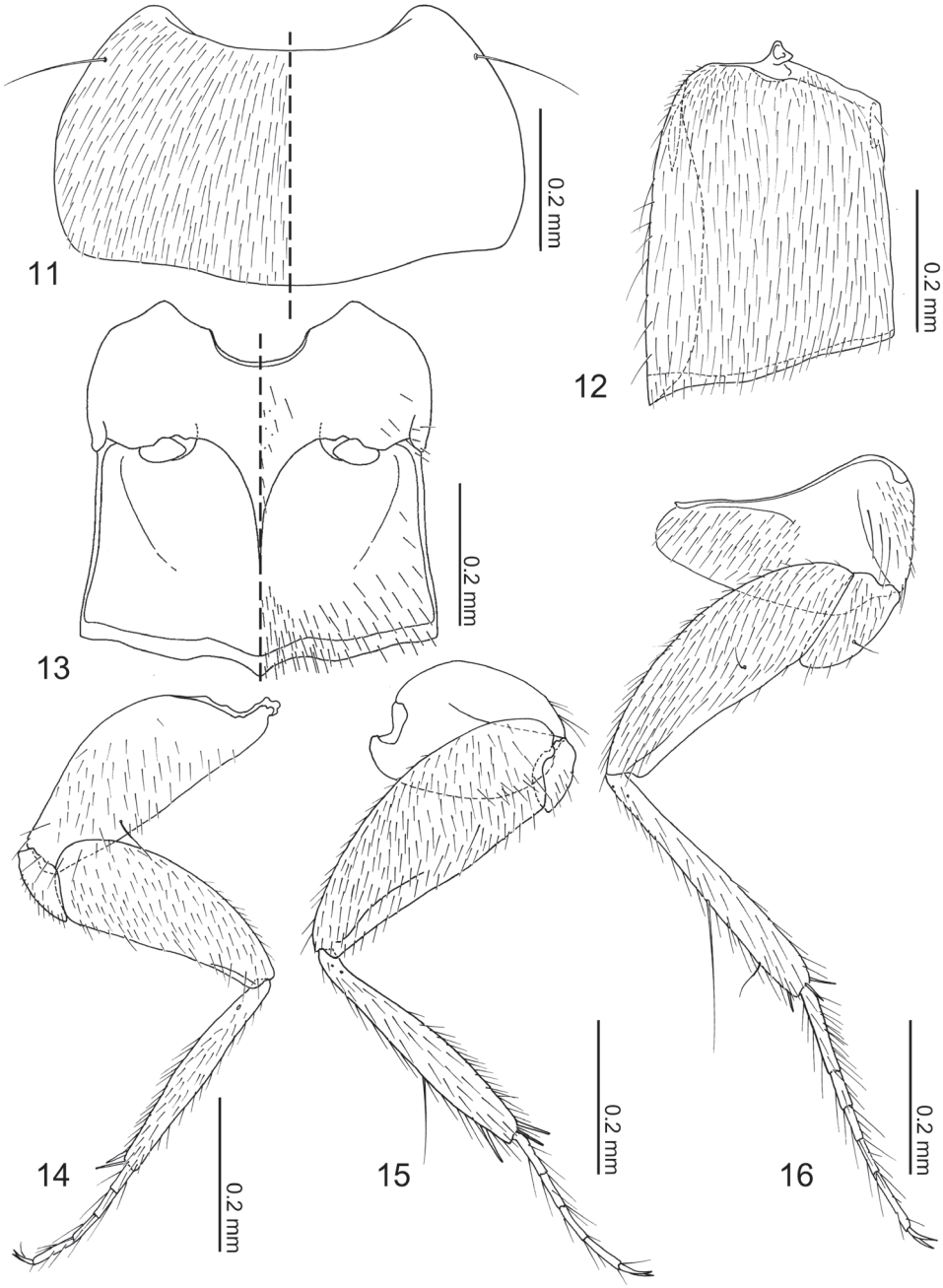
Median lobe of aedeagus (Figs 20–21) with basal capsule bulbous, more than three times as wide as apical lobe (Fig. 20), paramerally expanded (Fig. 21). Paramere (Fig. 22) with a structure appearing homologous to Seevers' (1978) medial phragma produced apically; apical lobe extremely elongate and tapered apically, subequal in length to paramerite, single seta present. Spermatheca (Fig. 23) long and coiled.

Etymology. The generic name is derived from a combination of *Neo*, Greek for "new" and the name of a similar-looking genus *Termitosocius* Seevers, 1941. The gender is masculine.

Neotermitosocius bolivianus **Kanao, Eldredge & Maruyama, sp. n.**
urn:lsid:zoobank.org:act:1625FEFE-B75F-4C81-A9F9-65DAA629F1E5
http://species-id.net/wiki/Neotermitosocius_bolivianus
Figures 1–23

Type material. *Holotype*: ♂, "BOLIVIA: Cochabamba/117 km E Yungas/(Cochabamba-Villa Tunari Rd)/17°6'32"S; 65°41'12"W/ca. 1,040 m alt., 1–6-II-1999; R. Hanley ex. flight/intercept trap BOL1H99 028//SMO754884/KUNHM-ENT".

Paratypes: 2♂♂, ♀, same date and locality data as holotype (♂, SEMC barcode number "SMO75489", completely disarticulated; ♂, SEMC barcode number "SMO75488", abdominal segments VII–X dissected off; ♀, SEMC barcode number "SMO754880", head and abdominal segments VIII–X dissected, spermatheca not recovered). ♂, ♀, ?, same locality data as the holotype, differing data reads "8–10-II-1999", "BOL1H99 062" (♂, SEMC barcode number "SMO 754864", abdominal segments VIII–X dissected off; ♀, SEMC barcode number "SMO754858", completely disarticulated; ♀, SEMC barcode number "SMO754860", right antennomeres VI–XI and abdominal segments VIII–X dissected off; ♀, SEMC barcode number "SMO754863" and additional label reading "Termitohospitini/new genus/det. K. T. Eldredge 2011", abdominal segments VIII–X dissected off; ?, SEMC barcode number "SMO754857", hind wings removed); ♂, "BOLIVIA: Cochabamba/117 km E.



Figures 11–16. *Neotermitosocius bolivianus*, thorax and legs. **11** pronotum, left side with setae **12** elytron, left **13** meso- and metaventrites, anatomical left side with setae **14** fore leg, posterior surface **15** mid leg, anterior surface **16** hind leg, anterior surface.

Cochabamba, ai/Lagunitas, 1000m, 17°6'22"S/65°40'57"W, 6–8-II-1999/F. Genier, mountain evergreen/forest, ex. flight intercept trap/99-037// SMO754908/KUNHM-ENT", completely disarticulated.

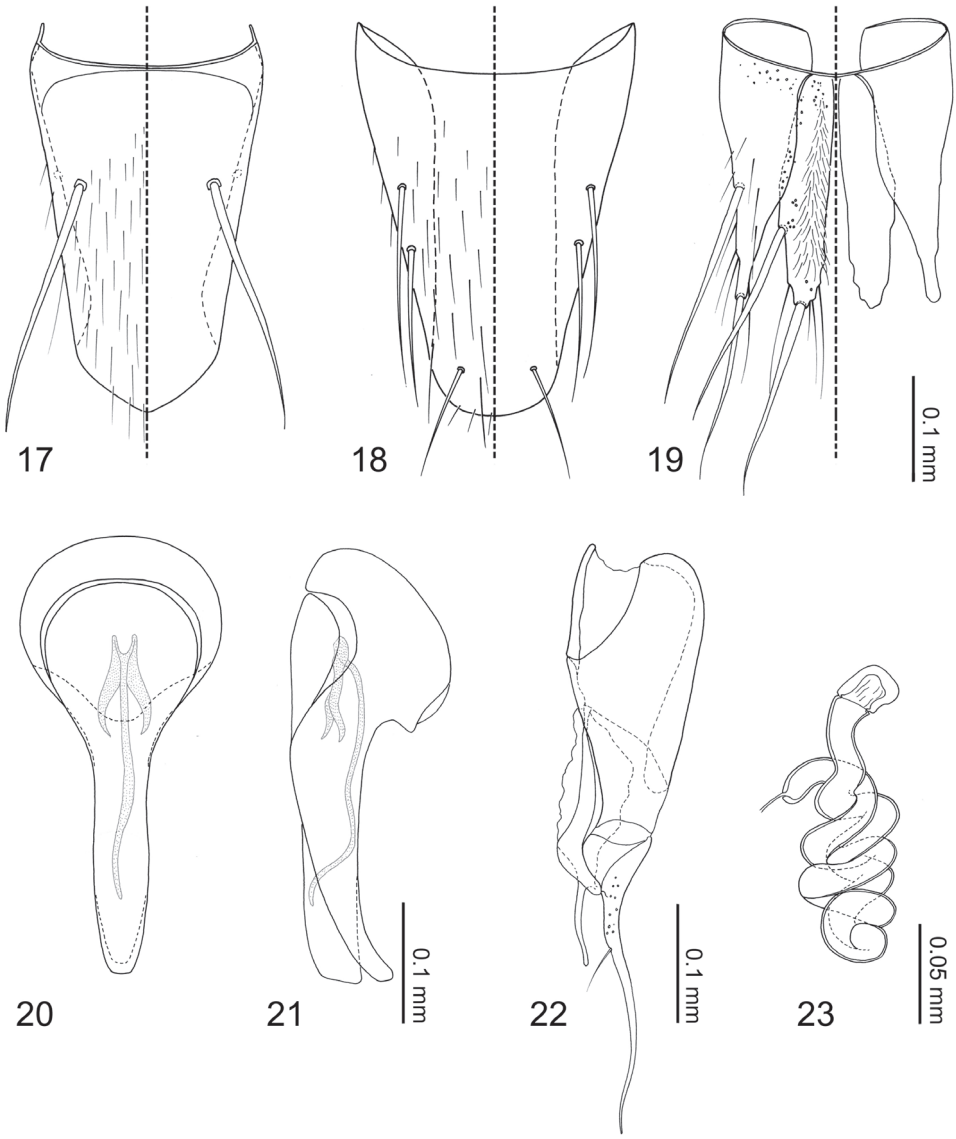
All type specimens are deposited in the Snow Entomological Museum Collection (SEMC).

Diagnosis. This species is diagnosable based on the generic diagnosis above.

Description. Body (Figs 1–2) approximately 2 mm in length (1.84–2.00 mm, N = 2), almost uniformly orange brown, but head slightly darker. Dorsal surface of head (Fig. 3) sparsely covered with setae, a pair of long setae present at anterior margin of clypeus. Eyes with uniform cover of inter-ommatidial setae. Antennae (Fig. 4) sparsely covered with setae; antennomere I with a long macroseta at middle of internal lateral surface; antennomere II with 4–5 macrosetae around apical margin, one macroseta conspicuously longer; antennomere III with 4 thick, very long macrosetae at apical margin; antennomeres IV–X with 3–4 macrosetae around apical margin; antennomere XI with several macrosetae near middle and apex. Labrum (Fig. 5) dorsal surface with 2 pairs of setae at anterolateral corners, 3 pairs of longer and 3 pairs of shorter setae at mesal area of disc; epipharynx (Fig. 5) with two pairs of lateral setulae. Mandible (Figs 6–7) dorsum with 2 pairs of scrobal setae near middle of aboral margin, six setae around base of disc. Maxillary (Fig. 8) lacinia with 2 pores at middle; galea with 3 pores apicomedially; maxillary palpal article I with a long seta at lateral margin and a medial pore, article II sparsely covered with setae, article III sparsely covered with setae and 2 stronger setae medially.

Pronotum (Fig. 11) transverse (pronotum length = 0.30–0.40 mm, pronotum width = 0.54–0.72 mm, N = 7), densely covered with setae, with a pair of long macrosetae at anterolateral corners. Elytra (Fig. 12) longer than wide (elytra length = 0.42–0.44 mm, elytra width = 0.30–0.34 mm, N = 5), densely covered with setae, setae of lateral margins longer. Mesoventrite (Fig. 13) sparsely covered with minute setae at central and posterolateral areas. Metaventrite (Fig.13) posterior one third sparsely covered with setae. Fore leg (Fig. 14) uniformly covered with setae; coxa with a macroseta near dorsomedial of anterior surface and 5–6 long setae at apical margin; femur with longer setae near anterior inner margin; tibia with apical setae stronger, 2 spurs present at apex. Mid leg (Fig. 15) uniformly covered with setae except coxa; coxa with several long setae around anterior margin; tibia with long macroseta at middle on dorsal surface, 3 spurs present at apex. Hind leg (Fig. 16) uniformly covered with setae, coxa medially nude; coxa with 3 longer setae along cavity; trochanter and femur with a macroseta near middle of anterior surface; tibia with one longer and shorter macroseta on anterior surface, 3 apical spurs present.

Tergites and paratergites III–VIII (Figs 1, 17) sparsely covered with setae. Macrochaetotaxy of abdominal tergites III–VIII = 0-0-2-2-0-2. Sternite VIII (Fig. 18) sparsely covered with setae; 3 pairs of macrosetae present, 2 pairs at mediolateral margin and one near apex. Tergite IX (Fig. 19) with pores around anterior margin, several setae near middle of disc, 2 pairs of macrosetae at mediolateral margin and apex present. Tergite X (Fig. 19) covered in setae, 4 longer setae at apex, 2 pairs of macrosetae present at mediolateral margin and apex.



Figures 17–23. *Neotermitosocius bolivianus*, abdominal sclerites and genitalia. **17** tergite VIII **18** sternite VIII **19** tergites IX–X **20** median lobe of aedeagus, aparameral view **21** median lobe of aedeagus, lateral view **22** paramere, external view **23** spermatheca.

Male. Median lobe of aedeagus (Figs 20 & 21) copulatory piece flagellate, suspensoria associated with lateral base of copulatory piece. Paramere (Fig. 22) apical lobe with pores at base.

Female. Spermatheca (Fig. 23) cuticle at apex with wrinkle-like sculpture.

Etymology. The specific epithet is derived from its type locality of Bolivia Latinized. The gender is masculine.

Distribution. Cochabamba, Bolivia.

Ecology. All known specimens of *Neotermitosocius bolivianus* were collected with a passive sample method (flight intercept trap) and nothing is known about its biology. It is hypothesized to be a termite symbiont based on phylogenetic relationships but the host is unknown.

Coptotermocola Kanao, Eldredge & Maruyama, gen. n.

urn:lsid:zoobank.org:act:01A98C55-BA86-4064-A554-1EC1DA6DC93A
http://species-id.net/wiki/Coptotermocola

Type species: *Coptotermocola clavicornis* sp. n.

Diagnosis. This monotypic genus is distinguishable from all other Termitohospitini most distinctly by the strongly carinate mesocoxal process (Figs 27, 38), and additionally by the compact antennae (Fig. 29) and short maxillary palpi (Fig. 33).

Description. Body form (Figs 24–27) limuloid. Pronotum strongly convex, dorsally obscuring most of head; abdomen posteriorly tapered.

Head capsule (Fig. 28) transverse, widest behind eyes. Antennal fossae deep, as large as eyes and dorsally obscured by vertex. Eyes large and produced anterolaterally. Antennae (Fig. 29) compact with 11 articles; antennomere I longer than II–X and partially hidden within antennal fossae; antennomere II dilated at apex to receive antennomere III; antennomeres III–X strongly transverse and more than twice as wide as long, length increasing distally; antennomere XI fusiform; pedicles of antenomeres IV–XI obscured by apex of preceding article. Labrum (Fig. 30) transverse and semicircular, anterior margin broadly concave; disc basomedially sparsely covered with pores; epipharynx (Fig. 30) glabrous. Mandibles (Figs 31–32) asymmetrical, dorsally covered with numerous pores around middle. Right mandible (Fig. 32) with a small subapical tooth; anterior one fifth strongly curved adorally. Left mandible (Fig. 31) without a subapical tooth. Maxillary (Fig. 33) lacinia elongate and strongly recurved adorally, adoral margin with 11 setae; apical two setae without basal cuticular processes (spinose scale of Ahn and Ashe 2004), third apical seta with a proximal basal cuticular process, central five setae with paired basal cuticular processes, basal three setae without basal cuticular processes; galea tapered apically, apex densely furnished with long trichae; palpus with 4 articles; article I triangular; article II trapezoidal and dilated toward apex; article III oviform, more than three times longer than wide and sparsely covered with long setae; article IV slightly narrowed toward apex with inconspicuous pseudosegment, pseudopores present apicomedially. Mentum (Fig. 34) trapezoidal, disc covered with long setae and several pores near middle and lateral areas; anterolateral corners slightly produced and with a pair of long macrosetae. Labial (Fig 35) palpus with 3 articles, first article a fusion of I + II; article I + II with a mediobasal seta on ventral surface (probably homologous to seta c of Sawada [1972]), twin and medial pores present; article III almost half as long as article I + II with 2 pores at apex; ligula triangular with medial subtriangular area sclerotized; prementum wider than long, with 2 pairs of both

Figures 24–27. Habitus of *Coptotermocola clavicornis*. **24** beetle photographed *in situ* **25** dorsal view **26** lateral view **27** ventral view.

real and pseudopores, and a pair of setal pores present; apodemes broad and longer than disc, dilated posteriorly with apices recurved internally and almost touching.

Pronotum (Fig. 36) transverse, widest at posterolateral angles; anterior margin concave with anterolateral angles slightly produced anteriorly; posterior margin rounded and slightly produced medially; marginal cuticle thin and somewhat translucent (Fig. 26). Prosternum reduced in length. Elytra (Fig. 37) subquadrate, slightly wider than long. Wings fully developed. Mesoventrite (Fig. 38) slightly shorter than metaventrite;

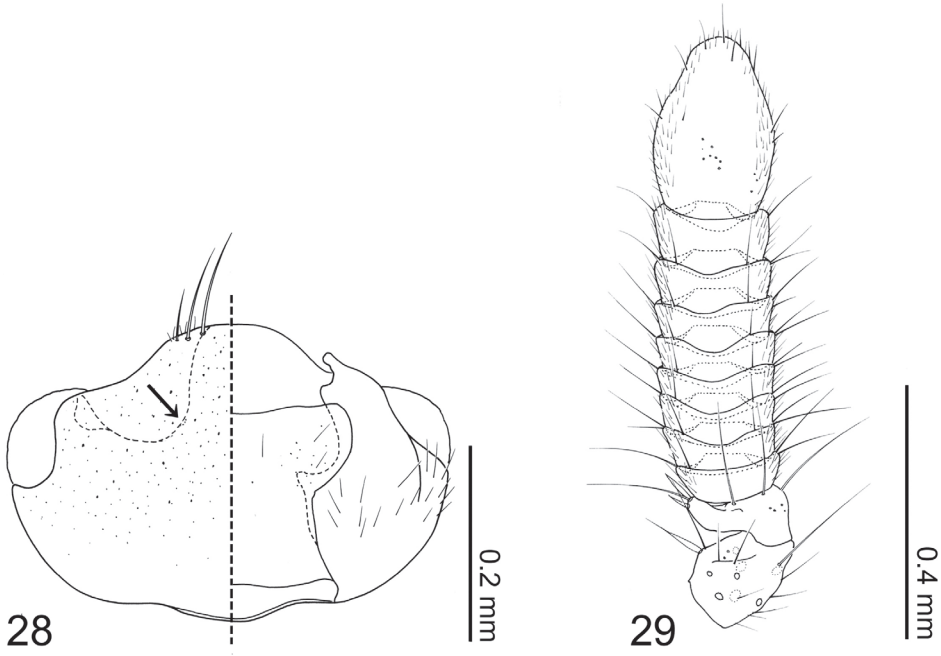


Figure 28–29. *Coptotermocola clavicornis*. **28** head capsule left side = dorsal view, right side = ventral view **29** antenna.

mesoventral process produced as an extremely large structure widest medially and tapered at ends, microsculpture composed of longitudinal wavy lines and pores (Fig. 39), structure ventrally partially obscuring basal podomeres; mesocoxal cavity marginal bead complete, narrowly separated. Legs (Figs 40–42) stout and laterally flattened; femora subrectangular; apical tarsomeres longest, tarsal formula 4-4-5. Fore leg (Fig. 40) with coxa approximately as long as femur; trochanter subtriangular; tibia thin. Mid leg (Fig. 41) with coxa globular; trochanter very thin; tibia thin. Hind leg (Fig. 42) subrectangular and almost as long as trochanter and femur combined; trochanter globular-triangular; femur slightly dilated apically; tibia thin.

Abdomen (Figs 24–27) tapered posteriorly. Segment I represented by only tergite I fused to the metanotum. Segment II represented by only by tergite II. Segments III–VII with 1 tergite, 1 sternite, and 2 pairs of paratergites. Tergite VIII (Fig. 43) with a blunt apicomedial point. Sternite VIII (Fig. 44) with posterior margin rounded. Tergite IX (Fig. 45) fully subdivided dorsally by tergite X. Tergite X (Fig. 45) fully divided medially to base, only connected each other and tergite IX on anterior margin.

Median lobe of aedeagus (Figs 46–47) with basal capsule bulbous. Paramere (Fig. 48) with a structure appearing homologous to Seevers' (1978) medial phragma produced apically. Spermatheca (Fig. 49) apically slightly bulbous.

Etymology. The generic name is derived from a combination of the generic name of the host termite, *Coptotermes* Wasmann, 1896 and the Latin noun *cola* meaning "dweller". The gender is feminine.

Coptotermocola clavicornis **Kanao, Eldredge & Maruyama, sp. n.**
urn:lsid:zoobank.org:act:FEF68FC3-CB00-4B41-95C8-A6DA28E8FFA7
http://species-id.net/wiki/Coptotermocola_clavicornis
Figures 24–49

Type material. *Holotype*: ♂, "MALAYSIA: Selangor,/Ulu Gombak, 03°19'479"N; 101°45'170"E,/ca. 240 m alt., X July 2011,/T. Kanao leg. KT-261". Abdominal segments VIII–X dissected off.

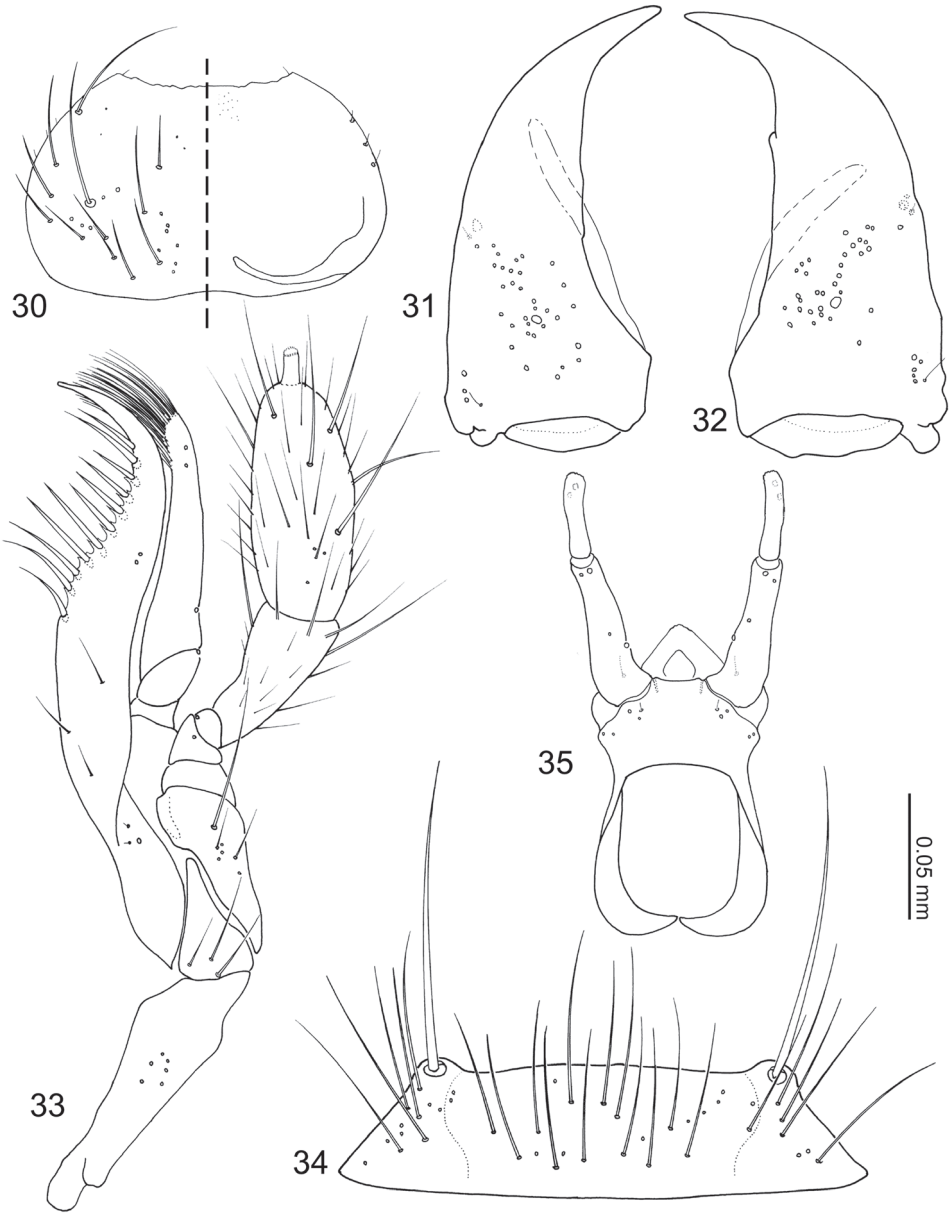
Paratypes: 6??, MALAYSIA: same data as the holotype, one specimen is preserved in 99.5% EtOH; ♀, same locality data as the holotype, differing data reads "XXI May 2010,/ T. Kanao leg. KT-33", fully disarticulated; 3??, same locality data as the holotype, differing data reads "XXIX May 2012,/ T. Kanao leg. KT-312", one specimen is preserved in 99.5% EtOH.

All type specimens are deposited in the Kyushu University Museum.

Diagnosis. This species is diagnosable based on the generic diagnosis above.

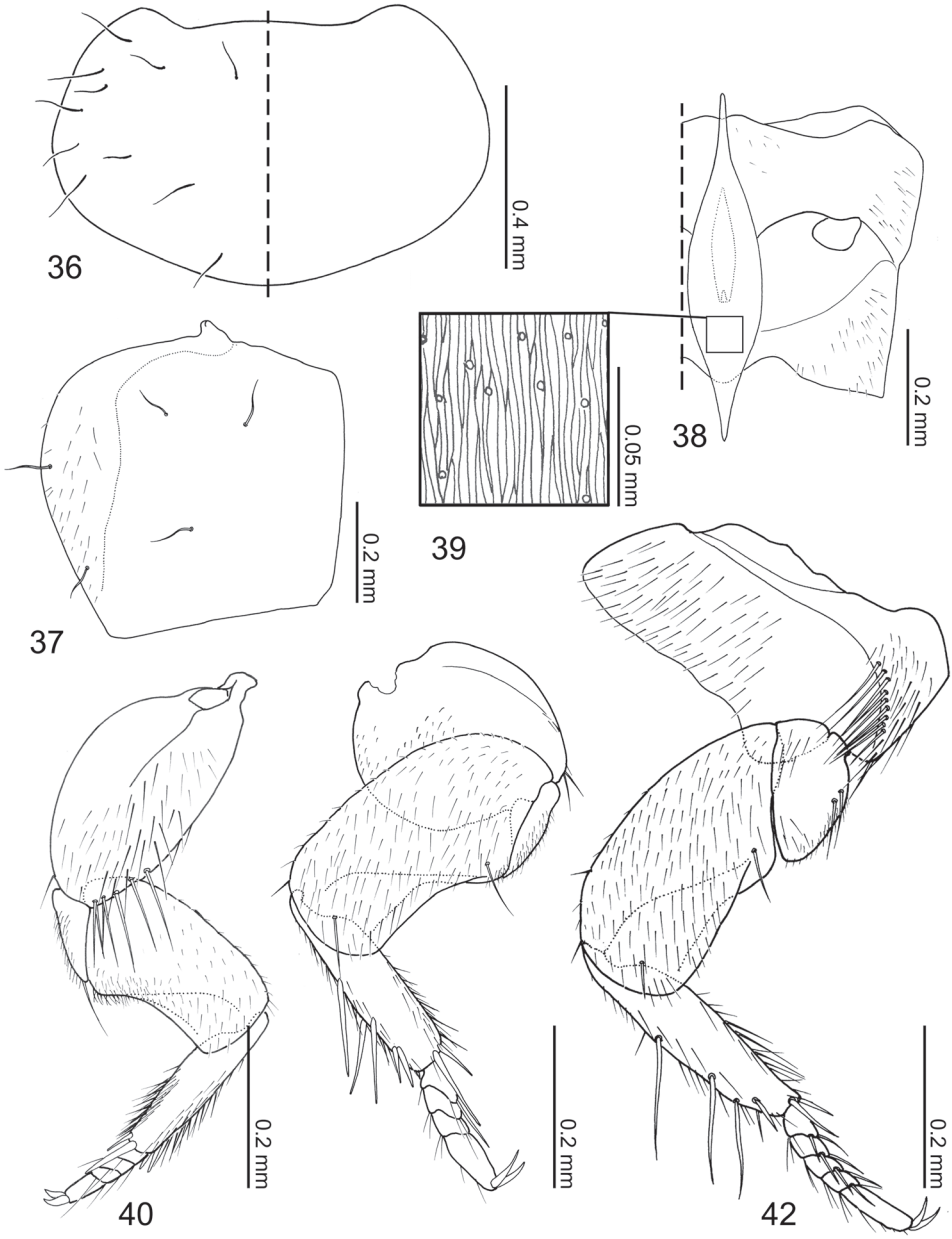
Description. Body (Figs 24–27) approximately 2 mm in length (1.71–2.16 mm, N = 4) almost uniformly reddish brown, but head slightly darker. Dorsal surface of head (Fig. 28) glabrous, sparsely covered with pores, with 3 pairs of long setae at anterior margin of clypeus; ventral surface (Fig. 28) with several setae behind eyes. Antennomere (Fig. 29) I sparsely covered with pseudopores and several macrosetae; antennomere II with 6–7 long macrosetae, 2 of them stronger and several pores present; antennomeres III–X sparsely covered with setae and 3–4 macrosetae present; antennomere XI sparsely covered with setae, with several macrosetae on dorsal and ventral surface near apex, pores present centrally on lateral surface. Labral (Fig. 30) surface with 14–16 setae, anterolateral marginal and near-middle pairs conspicuously stronger. Epipharynx (Fig. 30) with a pair of setulae present on anterolateral corner and three pairs of lateral marginal setulae. Mandibles (Figs 31–32) with seta present at aboral basolateral margin. Maxillary (Fig. 33) lacinia mesally with two pores and basally with 3 setae present; galea with 2 pores apically; maxillary palpal article I with a medial pore, article II sparsely covered with setae and longer setae present on apical margin, article III sparsely covered with longer and shorter setae.

Pronotum (Fig. 36) transverse (pronotum length = 0.55–0.62 mm, pronotum width = 0.91–1.02 mm, N = 6) with 11 pairs of macrosetae. Elytra (Fig. 37) subquadrate (elytra length = 0.50–0.60 mm, elytra width = 0.51–0.63, N = 6), disc laterally sparsely setose, 2 lateral and 3 discal pairs of macrosetae present. Mesoventrite (Fig. 38) with central and lateral setose areas. Metaventrite (Fig. 38) with posterolateral setose area. Fore leg (Fig. 40) with coxa sparsely setose and 5 long macrosetae present at apical margin; trochanter and femur sparsely covered with setae; tibia covered with setae, density increasing apically, 5 apical spurs present; tarsus with few setae. Mid leg (Fig. 41) with coxa sparsely setose, 2 macrosetae at apex; trochanter sparsely covered with setae; femur overall setose, macroseta present venterobasally; tibia covered with setae, density increasing apically, with 7 strong setae present dorsally and apically



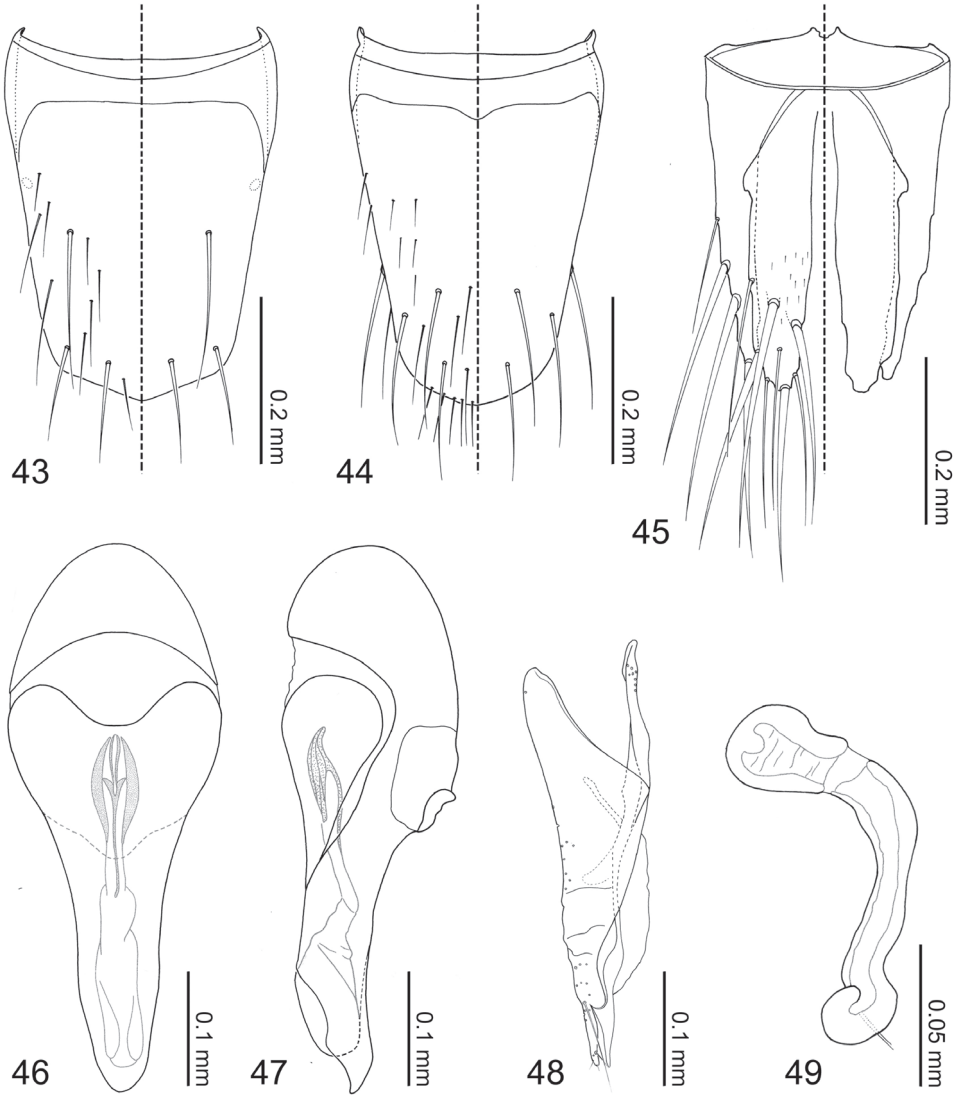
Figures 30–35. *Coptotermocola clavicornis*, mouthparts. **30** labrum, left side = labrum, right side = epipharynx **31** left mandible, dorsal view **32** right mandible, dorsal view **33** maxilla, ventral view **34** mentum, ventral view **35** labium, ventral view.

with basal three dorsal setae longest; tarsomeres with 3–4 setae at apical margin. Hind leg (Fig. 42) with coxa mostly setose and with approximately 10 macrosetae along femoral cavity margin; trochanter partially setose and with 2 macrosetae along ventrolateral margin; femur overall setose, one macroseta near base and three macrosetae



Figures 36–42. *Coptotermocola clavicornis*, thorax and legs. **36** pronotum, left side with and right side without setae **37** elytron, left **38** meso- and metaventrites, anatomical left side with setae **39** detail of mesosternal process surface sculpture **40** fore leg, posterior surface **41** mid leg, posterior surface **42** hind leg, posterior surface.

apically present; tibia sparsely covered with setae, with 7 strong setae present dorsally and apically, basal three dorsal setae longest; tarsomeres with 3–4 micro- and 2 macrosetae present at apical margin.



Figures 43–49. *Coptotermocola clavicornis*, abdominal sclerites and genitalia. **43** tergite VIII **44** sternite VIII **45** tergites IX–X **46** median lobe of aedeagus, aparameral view **47** median lobe of aedeagus, lateral view **48** paramere, external view **49** spermatheca.

Tergites III–VIII (Fig. 25, 43) laterally setose and medially glabrous. Macrochaetotaxy of abdominal tergites III–VIII = 2-4-4-4-4-4; paratergites setose. Tergite VIII (Fig. 43) with a pair of discal and two pairs of apical macroseta present. Sternite VIII (Fig. 44) sparsely setose and with one discal and three marginal macrosetae. Tergite IX (Fig. 45) with 3 pairs of macrosetae at apex and lateral margin; tergite X (Fig. 45) disc sparsely covered with minute setae and 4 pair of macrosetae near apex.

Male. Median lobe of aedeagus (Figs 46–47) copulatory piece flagellate, suspensoria associated with lateral base of copulatory piece. Paramere (Fig. 48) condylite with pores basally; apical lobe with 4 setae present.

Female. Spermatheca (Fig. 49) apical bulb surface with transverse wrinkle-like sculpture; stalk basal to membranous area three times as long as apical bulb.

Etymology. The specific epithet is derived from a combination of the Latin noun *clava* meaning "club" and Latin adjective *cornis* meaning "to be horned", in reference to the diagnostic robust antennae of the species. The gender is feminine.

Distribution. Known only from the type locality Ulu Gombak, Selangor, Malaysia.

Host species. All specimens were collected from the nest of *Coptoptermes gestroi* (Wasmann, 1896). Ahmad (1965), Kirton and Brown (2003) and Tho (1992) was consulted for host identification.

Ecology. Specimens acquired during the KT261 collecting event were collected from the galleries of the host termites within a rotting log. The galleries were large and arranged in a complex manner. Another specimen (KT33) was collected from a trail of the hosts that occupied the exterior of a large log. KT312 specimens were collected from a rotting log occupied by the host termites. All *Coptotermes* colonies that yielded *C. clavicornis* were located near rivers where the habitat in general was comparatively more moist compared to its surroundings.

All *C. clavicornis* specimens moved faster than their host termites. They did not avoid contact with hosts but instead recurved their abdomens over their bodies when they came into contact. The inquilines wedged themselves under their hosts on several occasions, but the host termites regaurdless never attacked the beetles.

Acknowledgments

T. K. and M. M. would like to express our thanks to Dr. Rosli Hashim of the University of Malaya for supporting and provisioning our fieldwork. T. K. thanks Associate Professor Satoshi Kamitani for reviewing the manuscript. K. T. E. would like to personally thank Zachary Falin and Jennifer Thomas for their sincere help with specimens deposited at the Univeristy of Kansas, and Andrew Short for support. Finally, we would all like to thank Dr. Takashi Komatsu for assistance in the field and for permission to use the photograph used in Fig. 24. Finally, we all thank M. Gimmel for tremendously helpful comments on drafts.

Contribution from the Entomological Laboratory, Faculty of Agriculture, Kyushu University, Fukuoka (Ser. 6, No. 115).

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ZooKeys 254: 89–97 (2012)
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RESEARCH ARTICLE

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Termitotrox cupido sp. n. (Coleoptera, Scarabaeidae), a new termitophilous scarab species from the Indo-Chinese subregion, associated with *Hypotermes* termites

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Academic editor: *A. Frolov* | Received 11 November 2012 | Accepted 11 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:F7941461-E375-440D-B1A2-23C2EC8542D4

Citation: Maruyama M (2012) *Termitotrox cupido* sp. n. (Coleoptera, Scarabaeidae), a new termitophilous scarab species from the Indo-Chinese subregion, associated with *Hypotermes* termites. ZooKeys 254: 89–97. doi: 10.3897/zookeys.254.4285

Abstract

Termitotrox cupido **sp. n.** is described from Cambodia and represents the first discovery of *Termitotrox* Reichensperger, 1915 from the Indo-Chinese subregion of the Oriental region. The type series was collected from fungus garden cells of *Hypotermes makhamensis* Ahmad, 1965 (Isoptera, Termitidae, Macrotermitinae). *Hypotermes* Holmgren, 1917 was previously an unknown host of *Termitotrox* species. The new species is readily distinguished from all known congeners by having wing-shaped trichomes on the elytra and is most probably the world's smallest scarab, at 1.2 mm in length.

Keywords

Termitophily, Termitotroginae, Termitotrogini, new species, Indo-Chinese subregion, smallest scarab

Introduction

The scarab genus *Termitotrox* Reichensperger, 1915 is composed of ten blind, flightless species from the Ethiopian region (eight species) and the Indian subregion of the Oriental region (two species) (Krikken 2008). Wasmann (1918) originally established the tribe Termitotrogini in the subfamily Coprinae of Scarabaeidae to include

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this genus, but the rank of the taxon was treated differently by different authors from a distinct family of Scarabaeoidea (Termitotrogidae: Tangelder and Krikken 1982; Dellacasa 1988), a subfamily of Scarabaeidae (Termitotroginae: Smith 2006; Bouchard et al. 2011), to a tribe of Aphodiinae (Termitotrogini: Scholtz and Grebennikov 2006). Currently, the subfamily status or the tribal status is accepted, but it remains unstable until a precise phylogenetic position of *Termitotrox* within Scarabaeidae is investigated. Most species have been found in fungus gardens of the fungus-growing termite genera *Odontotermes* Holmgren, 1912 or *Protermes* Holmgren, 1910 (Isoptera, Termitidae, Macrotermitinae). Recently, I collected an extraordinary, undescribed species of *Termitotrox* in Cambodia which is far from the known localities of the genus in the Oriental region (India) and is the first discovery of the genus in the Indo-Chinese subregion. It was collected from fungus garden cells of *Hypotermes* Holmgren, 1917 (also Macrotermitinae), a previously unknown host. This paper describes it as a new species.

When Arrow (1920) described *Termitotrox minutus* (originally in the genus *Aphodiocopris* Arrow, 1920) from India, he stated "probably the smallest Lamellicorn beetle [= scarab] hitherto discovered". *Termitotrox minutus* is approximately 1.5 mm in length. Subsequently Pittino (2007) described *Microtermitodius atomus*, ranging from 1.4 to 1.7 mm, which reduced the size of the smallest known scarab still further. However, the new species is 1.2 mm long, and is therefore likely the world's smallest scarab found thus far.

Materials and methods

In August 2012, I visited Angkor Wat, Siem Reap, Cambodia, a typical tropical monsoon forest, for five days and worked a total of 15 hours collecting termitophiles associated with fungus-growing termites. I examined fungus gardens of the genera *Macrotermes* Holmgren, 1910 (four species), *Odontotermes* (three species), *Microtermes* Wasmann, 1902 (one species) and *Hypotermes* (one species). I found 25 *Termitotrox* beetles from fungus garden cells of *Hypotermes makhamensis* Ahmad, 1965 by examining more than 80 fungus gardens. The beetles were put in a killing tube (35 ml) with tissue paper and ethyl acetate; two hours later they were removed from the tube and kept in 80% ethanol. Twenty two specimens were dried and mounted for morphological observation, and remaining three are kept in 99% ethanol for future DNA extraction. Specimen photographs were taken using a Canon EOS 60D with a Canon MP-E 65 mm 1-5× macro lens and mounted using a software CombineZM. Images of living beetle were taken by the same camera set in the field by Dr. Takashi Komatsu. Terminology of the species description follows Krikken (2008). The type series is deposited in the author's collection in the Kyushu University Museum, Fukuoka (KUM); Canadian Museum of Nature, Ottawa (CMN); The Natural History Museum, London (NHM); and the University of Nebraska State Museum (UNSM).

Taxonomy

Genus *Termitotrox* Reichensperger

Termitotrox Reichensperger 1915: 16 (type species: *Termitotrox consobrinus* Reichensperger, 1915, by monotypy).
Aphodiocopris Arrow 1920: 432 (type species: *Aphodiocopris minutus* Arrow, 1920, by monotypy).

Comments. See Krikken (2008) for generic review.

Termitotrox cupido Maruyama, sp. n.

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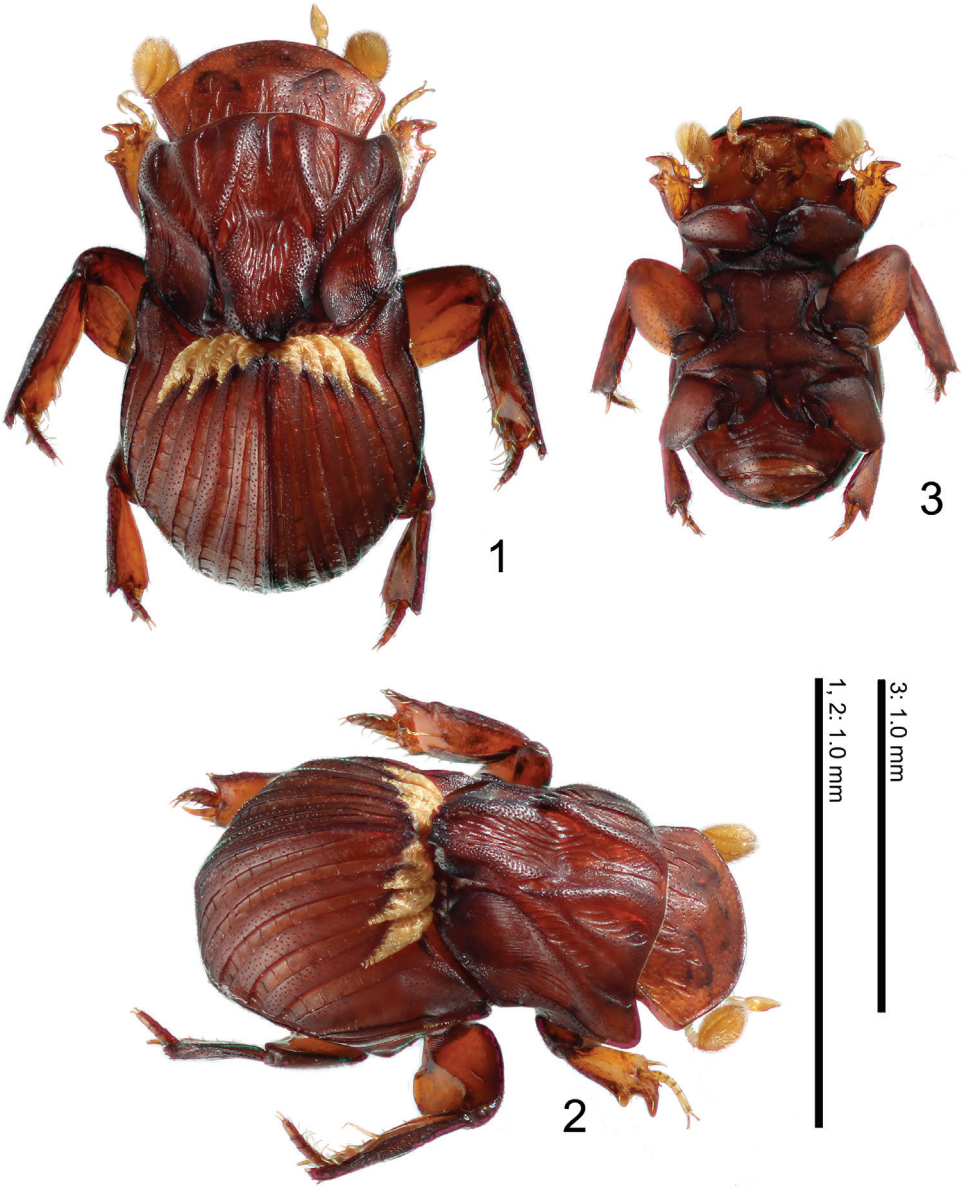
http://species-id.net/wiki/Termitotrox_cupido

Type materials. Holotype female, north of Preah Khan, Siem Reap, Cambodia, 19 VIII 2012, M. Maruyama (KUM). Paratypes, 6 males, 10 females, same data as holotype (KUM, NHM); 4 males, 1 females, 3 unsexed, same data but 21 VIII 2012 (CMN, KUM, UNSM).

Etymology. *Cupido* is the god of desire and love in Roman mythology and is often illustrated as a small, winged boy. The new species is named in reference to the wing-shaped trichomes on the elytra and the remarkably small body size. Noun in apposition.

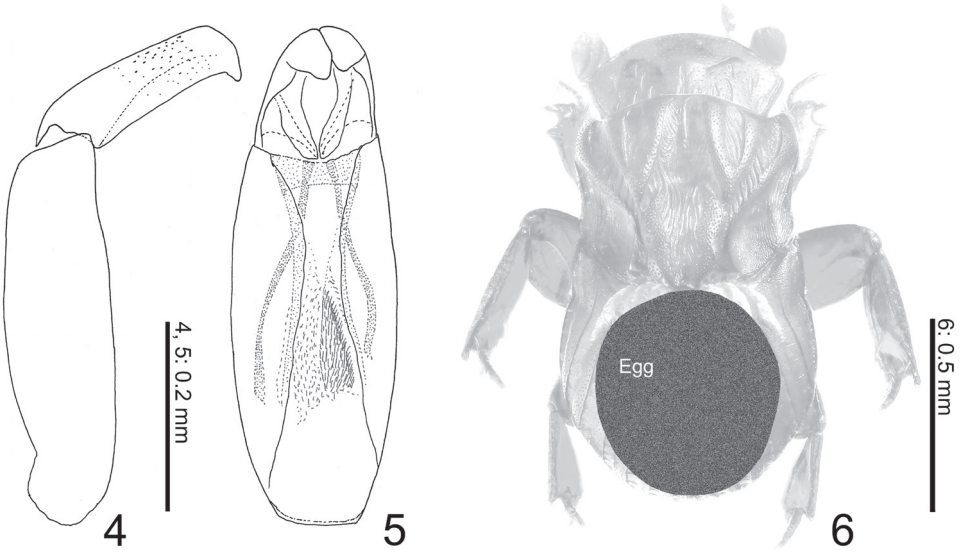
Diagnosis. This species is probably related to *T. minutus* (Arrow, 1920) because of its small body size and shape of elytra but easily distinguished from it by the spherical elytra, the presence of the trichomes on the elytra and the smaller body.

Description of holotype female. General colour uniformly reddish brown, slightly matt; length 1.21 mm. ***Head.*** Surface generally evenly convex, with only a slight callosity at clypeofrontal transition. Lateral margin of head entirely, finely marginate. Clypeal outline evenly rounded over entire length. Clypeofrons reddish brown, glabrous, distinctly, moderately punctate; vertex with deep groove medially, and 6 or 7 pairs of sharply defined, elongate primary punctures. Clypeofrontal border at (vague) suture straight; genal tip obtusely angular (in dorsal view); genal surface depressed. Antennal club yellowish brown. ***Prothorax.*** Prothorax reddish brown, narrower than elytra, sides (in dorsal view) evenly rounded over anterior half. Emargination at center apex not margined, anterolateral lobe rounded, edge slightly projecting downward (forming side of anterolateral propectoral cavity). Pronotal sides steeply declivous. Posterolateral section of pronotum rounded. Basolateral areas concave, with 1 feeble ridge around base; asymmetrical, left part with a tubercule near base. Apical lobe asymmetrical, with right side near apex roundly emarginate. Pronotal surface glabrous. Costae densely punctate, broader intercostal sulci with distinct, longitudinal wrinkles. Discal depression deep; surface, apart from some local micropunctation, smooth. Pronotal pattern of longitudinal costae as follows: Median costa indistinct around



Figures 1–3. Habitus of *Termitotrox cupido* sp. n. **1** Holotype, dorsal view **2** same, antero-lateral view **3** paratype, ventral view

apical 1/5; basomedian section narrow, surface deplanate, shallowly concave. Central depression posterolaterally delimited by depressed area of paramedian costa. Paramedian costa anteriorly broad, distinct, continuing to about half of pronotal length. Sublateral costa narrow, distinct, tapering posteriad to about half of pronotal length, reaching paramedian costa. Lateral costa anteriorly broad, distinct, extending from



Figures 4–6. *Termitotrox cupido* sp. n. **4** Aedeagus, lateral view **5** same, ventral view **6** mature egg (shaded circle) inside of female body.

anterolateral lobe caudad, tapering to base of pronotum. Marginal costa posteriorly broad, ending at depressed basolateral area. ***Elytra.*** Semi-spherical, reddish brown, matt, with 6 interstrial costae and intervening striae, and with trichomes at base of costae 2–6 to form wing-shaped patches. Humeral and apical elytral umbones absent; apicosutural edge nearly rectangular, slightly protruding. Epipleuron wide. Elytral striae distinct, deeply impressed, with transverse, weak costae from base to apex to form quadrate cells; striae 1 and 2 reaching basal half. Discal interstrial costae broadly trapezoidal (in cross-section), surface with dense, scattered punctures. Elytral pattern of interstrial costae as follows: costa 1 (next to suture) narrow, shiny, almost rectilinear; costa 2 shiny, tapering in front, stopping at basal half. Costa 3 complete, slightly narrowed at middle. Costae 4–6 complete, strongly developed, Costa 7, 8 and 9 apparently fused together. Anterolateral part of propectus deeply excavate. Preprosternal apophysis distinct, with several setae. Remainder of propectus glabrous, dark reddish brown. Posterolateral area of propectus with some ridges and grooves. Postprosternal surface with small, shallow, median impression. Transverse mesometasternal groove between posterior edges of mesocoxae distinct. ***Mesothorax.*** Mesosternum with a pair of identical, question-mark shaped grooves bordering the mesocoxae ; mesosternal surface reddish brown, glabrous, flattened; anterior surface densely micropunctate. ***Metathorax.*** Metasternum evenly convex, glabrous, and with fine perimarginal groove all around; reddish brown, infuscate laterally. ***Abdomen.*** Venter with 5 visible sternites, all reddish brown, matt, without grooves, sparsely micropunctate. Pygidium reddish brown, glabrous, base broadly margined; surface generally convex; surface lacking distinct microsculpture, sparsely micropunctate. ***Legs.*** Procoxa protuberant. Profemur brown, underside glabrous, sparsely micropunctate; outline broadly ellip-

Figure 7. *Termitotrox cupido* sp. n. walking on a fungus garden cell.

tic, emarginate distally. Protibia pale brown, broad, with short setae, microsculpture weak; shape strongly complanate, with 2 external denticles, no basal serration; apex straight, transverse, with distinct apico-internal spine; internal side strongly dilated from slender base. Protarsus twice longer than width of tibial apex, slender, yellowish; segment 1 inserted in fine groove, as long as segments 2–4 combined. Mesocoxae reddish brown, widely separated, slightly divergent anteriad. Mesofemora brown, broadly elliptic in outline, distally emarginate, surface moderately micropunctate, glabrous. Mesotibiae reddish brown, with several setae, broad, dilated near base, nearly parallel-sided from basal half to apex, edges entire; tibial apex deeply emarginate, with pair of acuminate apico-internal spurs, external one long, slightly curved, internal one short, straight; upper side of mesotibiae with fine longitudinal ridge near outer edge, underside with fine sinuate ridge from base to apico-internal section; with long setae around apical 2/5. Metatibiae similar to mesotibiae, but gently dilated apicad, with apex shallowly emarginate. Meso- and metatarsi brown, compacted-complanate, segments 1–4 short. Length of inner apical spur of metatibia 1/4 of metatibia, reaching base of tarsal segment 5.

Male. Aedeagus (Figs 4, 5) large, half as long as body length; paramere half as long as phallobase, gently narrowed apicad, curved near truncate apex.

Variations. Apical lobe of pronotum variable in shape, sometimes symmetrical, gently rounded at apex. Costa 7 of elytra sometimes indistinct.

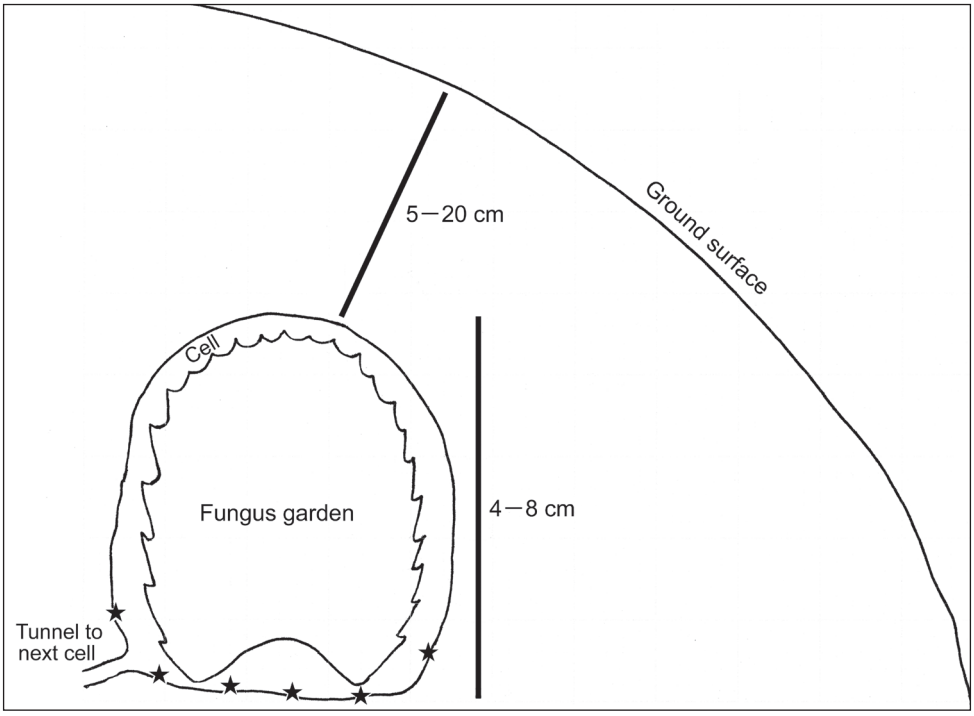


Figure 8. A schematic illustration of a fungus garden cell of *Hypotermes makhamensis* and places where *Termitotrox cupido* beetles were found (black stars).

Measurements in mm. Body length 1.13–1.22; maximum width of head 0.48–0.52; median dorsal length of pronotum 0.47–0.51, maximum width 0.54–0.59, sutural length of elytron 0.50–0.54, maximum width 0.62–0.67.

Symbiotic host. *Hypotermes makhamensis* (determined by Dr. Yoko Takematsu).

Remarks. No significant sexual dimorphism is detected. Male aedeagus is large compared with its body size. Female ovary contained a single huge egg occupying the greater part of the abdomen and metathorax (Fig. 6).

Discussion

Termitotrox cupido specimens were found only on the walls of the fungus garden cells of *Hypotermes makhamensis* (Figs 7, 8), unlike many other termitophiles associated with fungus-growing termites, which are usually found inside the fungus gardens themselves. In the same habitat (on the cell wall), undescribed species of *Odontoxenia* Schmitz, 1915, *Clitelloxenia* Kemner, 1932, *Ridiculiphora* Disney, 1997 (Diptera, Phoridae) and *Discoxenus* Wasmann, 1904 (Coleoptera, Staphylinidae) were found, but they were also found inside the fungus gardens.

When the fungus garden was removed from the cell, the beetles walked slowly on the cell wall to escape to a tunnel connected to the other fungus garden cells, by following the termites. No direct contact with the beetle by the termites was observed even though the beetle walked among highly dense columns of the termites. Since *Termitotrox cupido* is so small (≈ 1.2 mm) and the termites were rushing to escape the disturbance, further observations of its termite association were not possible.

No behavioural information of *Termitotrox* species is available, other than that reported here for *T. cupido*. In two species of Corythoderini, the other termitophilous scarab tribe associated with fungus growing termites, beetles were being carried by worker termites as they do for their own nymphs (Kistner 1982; Maruyama 2012): the worker termite grabbed a handgrip-like structure which is formed by the median lobe of the pronotal base (medially projected posterior margin) and a median projection of the elytral base of the beetles. Though *Termitotrox* species do not possess an elytral median projection, they share the pronotal median lobe which is well developed and is handgrip-shaped, hence it could be hypothesized that *Termitotrox* beetles are likewise carried by the worker termites.

Termitotrox cupido is characterized by the large trichomes on the elytral costae, and these trichomes are not known among other *Termitotrox* species. Among inquilinous Aphodiinae trichomes on body surfaces have been observed in members of the tribes Corythoderini, Termitoderini, Stereomerini, and Rhyparini. Wasmann (1903) observed secretory tissue in *Chaetopisthes assmuthi* Wasmann 1911 (Corythoderini) under the posterior part of the pronotum and apices of the elytra, where the trichomes originate. This kind of tissue could exist in the trichomes of other species, and it probably produces chemical signals that target the host termites. Krikken (2008) speculated that the body surface convexities (including elytral costae) in the other *Termitotrox* species that do not possess trichomes might be filled with glandular tissue. The trichomes on the elytral costae in *T. cupido* indicate the presence of glandular tissue in the convexities of the elytral costae, suggesting glands might by present in this body region in the other species.

In the Oriental region the known distribution of *Termitotrox* species was restricted to the Indian subregion before the present finding of *T. cupido* in Cambodia in the Indo-Chinese subregion. Krikken (2008) mentioned that the Indo-Afrotropical distribution of *Termitotrox* is analogous to that of the Corythoderini. Recently, I collected a new genus and species of Corythoderini in the same place as *T. cupido* in Cambodia (Maruyama 2012). Aanen & Eggleton (2005) suggested that fungus-growing termites originated in the African rainforest, and their distributional range subsequently extended from tropical Africa to Southeast Asia. Corythoderini and *Termitotrox* might also have originated in Africa and have extended their distributions along with their host termites.

Acknowledgments

I thank Dr Yoko Takematsu (Yamaguchi University) for identification of the termite host; Dr Takashi Komatsu (Shinshu University) for taking the pictures of the living individual (Fig. 7); Mr Chamroeun Pisyth (Siem Reap) for assistance in the field; Dr

Brett Ratcliffe (University of Nebraska State Museum) for critically reading the manuscript and for information on the smallest scarab; Dr. Alberto Ballerio (Brescia, Italy), Dr. Andrey Frolov (Zoological Institute of Russian Academy of Sciences) and Dr. Joseph Parker (Columbia University) for reviewing the manuscript.

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ZooKeys 254: 99–108 (2012)
doi: 10.3897/zookeys.254.4182
www.zookeys.org

RESEARCH ARTICLE

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A taxonomic study on the genus *Ettchellsia* Cameron, with descriptions of three new species (Hymenoptera, Megalyridae, Dinapsini)

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Academic editor: *C. van Achterberg* | Received 25 October 2012 | Accepted 11 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:D4519DDA-6E5F-43DA-8876-FE0434A305E5

Citation: Mita T, Shaw SR (2012) A taxonomic study on the genus *Ettchellsia* Cameron, with descriptions of three new species (Hymenoptera, Megalyridae, Dinapsini). ZooKeys 254: 99–108. doi: 10.3897/zookeys.254.4182

Abstract

Three new species of *Ettchellsia* Cameron, namely, *E. ignita* **sp. n.** from Peninsular Malaysia and Borneo, *E. nigripes* **sp. n.** from Sulawesi and *E. reidi* **sp. n.** from Borneo are described and illustrated. A key to the species of *Ettchellsia* is provided based on females.

Keywords

Taxonomy, parasitic wasps, South East Asia

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Introduction

Cameron (1909) described a remarkable new wasp genus from Borneo that he dedicated to his housekeeper, Mary Ettchells. Cameron found the insect so unusual that he struggled to place it to family, and debated if it should be placed into a new family. Subsequent authors (Baltazar 1961; Shaw 1988, 1990; Vilhelmsen et al. 2010) have clarified the placement of *Ettchellsia* Cameron in the family Megalyridae. *Ettchellsia* Cameron 1909 is a small genus comprising three described species (Cameron 1909; Baltazar 1961; He 1991). *Ettchellsia* species show an Indomalayan distribution, i.e., *E. piliceps* Cameron occurring in Borneo, *E. philippinensis* Baltazar occurring in Philippines and *E. sinica* occurring in Yunnan, China. Shaw (1990) defined *Ettchellsia* as a monophyletic genus based on three synapomorphies: the rugose hind tibia, unique pattern of propodeal carinae (as in Figs 10–12), and smooth posterior border of the mesopleuron, without a row of foveae separating the mesopleuron and metapleuron. Shaw (1990) also examined specimens from Thailand and Viet Nam, which he regarded as range extensions and variations of *Ettchellsia piliceps* Cameron. One specimen from Taiwan was not placed to species because it was a male. Vilhelmsen et al. (2010) also referred to unidentified species of *Ettchellsia* from Taiwan and Thailand. According to recent phylogenetic studies, the genus *Dinapsis* Waterson (distributed in the Afrotropical region) was strongly suggested to be the closest relative of *Ettchellsia* (Shaw 1990; Vilhelmsen et al. 2010). *Ettchellsia* species are quite rare; however, recently some specimens were obtained from Peninsular Malaysia, Sulawesi and Kalimantan. They were identified by us as three different new species. These findings and recent taxonomic works on the genus *Carminator* Shaw, the other genus of the family found in Asia (Mita et al 2007; Mita and Konishi 2011) may imply the megalyrid species diversity of the Indomayan region is still not clarified well. The biology of *Ettchellsia* has remained unknown, and no distributional areas are reported except for type localities. It is almost the same case with *Carminator*. Based on a knowledge of other Megalyridae, *Ettchellsia* is presumed to be an idiobiont ectoparasitoid, probably attacking beetle larvae (Shaw 1990). Southeast Asia is a key area to understanding the complicated evolutionary history of the family Megalyridae because the sister-group pairs of both *Ettchellsia* (*Dinapsis* Waterson: Afrotropical) and *Carminator* (*Cryptalyra* Shaw: Neotropical) occur in far separated areas (Shaw 1990; Vilhelmsen et al. 2010). Further findings from Southeast Asia will help the understanding of the evolutionary history of the Megalyridae.

Materials and methods

The morphological terms used in the descriptions follow Shaw and van Noort (2009) and Vilhelmsen et al. (2010). Photo images were produced using Leica Application Suite (Leica Microsystems) and Combine ZM software (Alan Hadley, www.hadleyweb.pwp.blueyonder.co.uk). Line drawings were made using a drawing tube attached to a

binocular microscope. The depositories of the types are as referenced after the collection data.

Taxonomy

Genus *Ettchellsia* Cameron, 1909
http://species-id.net/wiki/Ettchellsia

Ettchellsia: Cameron 1909: 208; Baltazar 1961: 219; He 1991: 475

Type species. *Ettchellsia piliceps* Cameron, 1909 (monotypic).

Diagnosis. Eye margined posteriorly by post-orbital orbital carina; posterior border of mesopleuron smooth, without row of foveae; propodeum bearing unique pattern of longitudinal carinae (Figs 12–14); fuscous banding pattern present on fore wing (Figs 1, 4, 7) or wing entirely fuscous; fore wing with RS between RS+M and r-rs tubular (Fig. 4); apical part of RS tubular, arched toward stigma (Fig. 4); M+Cu and Cu1 spectral (Fig. 4); hind coxa bearing longitudinal carina; hind tibia rugose (Figs 1, 4, 7).

Detail generic character states were discussed by Shaw (1990) and Vilhelmsen et al. (2010).

Key to the females of the species of *Ettchellsia*

1 Ocellar triangle smooth, without a longitudinal row of punctures; smooth crescent-shaped depression present on outer margin of each ocellus; wings mostly clear except with some infumation medially and apically, but lacking distinct dark bands..... ***E. philippinensis* Baltazar**

– Ocellar triangle sculptured with at least one median longitudinal row of punctures (Fig. 2); crescent-shaped depressions absent near ocelli, or depressions irregularly foveate, not smooth; wings with three or four distinct dark bands..... **2**

2 Vertex strongly flattened, appearing flat in lateral view (Fig. 11); mesonotum more weakly setose, setae not covering surface ..... ***E. piliceps* Cameron**

– Vertex well-developed and convex, appearing round in lateral view (Fig. 10); mesonotum medially and posteriorly with thick setae largely covering surface..... **3**

3 Mesoscutum strongly humped (Fig. 3) **4**

– Mesoscutum not humped (Fig. 6) **5**

4 Median region of propodeum anteriorly strongly narrowed (as in Fig. 12); hind tibia and basitarsi dorsally with long black erect setae, many of which are

longer than the width of the hind tibia; metasoma reddish brown (Fig. 1).... ..... ***E. ignita*** **Mita & Shaw, sp. n.**

– Median region of propodeum not anteriorly strongly narrowed; hind tibia and basitarsi dorsally with long erect white (not black) setae, most of which are shorter than the width of the hind tibia; metasoma black (as in Fig. 4) ... . ***E. sinica*** **He**

5 Median region of propodeum medially narrowed (Fig. 14); vertex between posterior ocellus and eye rugose (Fig. 8)..... ***E. reidi*** **Mita & Shaw, sp. n.**

– Median region of propodeum anteriorly weakly narrowed (Fig. 13); vertex between posterior ocellus and eye mostly smooth (Fig. 5)..... ..... ***E. nigripes*** **Mita & Shaw, sp. n.**

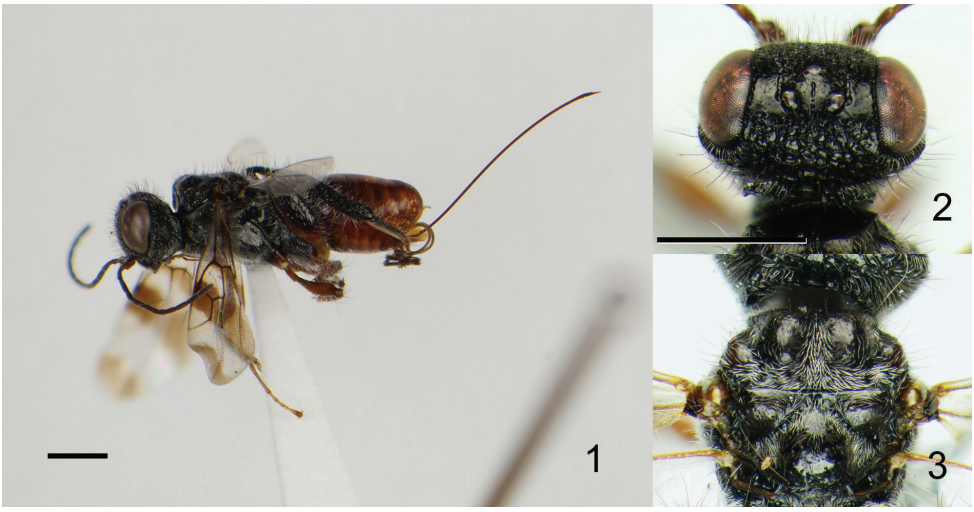
Ettchellsia ignita **Mita & Shaw, n. sp.**
urn:lsid:zoobank.org:act:5CDAC4E0-568D-4F4A-A72B-63AA5A4051C2
http://species-id.net/wiki/Ettchellsia_ignita
Figs 1–3, 10, 12

Type series. Holotype ♀: "MALAYSIA: Negeri Selangor, Ulu Gombak (Univ. Malaya Field Studies Centre, 220m alt) Malaise trap, 7–11.iv.2007 T. Tsuru & M. Maruyama leg.", "HOLOTYPE: *Ettchellsia ignita* Mita & Shaw, 2012, sp. n.". Paratype: 1♂, Sandan, Borneo, Baker leg. The holotype is deposited in the Systematic Entomology, Hokkaido University Museum, Sapporo, Japan (SEHU).The paratype is deposited in the U.S. National Museum of Natural History, Washington D.C, USA (NMNH).

Description. (Female) Head (Figs 1–2) 1.48 × wider than long, covered with long erect setae and short decumbent setae; frons reticulate; surface around ocellar triangle smooth with rows of punctures behind anterior ocellus and outside of posterior ocelli; vertex reticulate-rugose; eye margined posteriorly by foveate groove and single postocular orbital carina; gena sparsely punctate with irregular carinae under orbital carina; occipital carina forming a small depression; clypeus transversely rugose.

Mesosoma (Fig. 3) entirely covered with short decumbent white setae, but long erect setae also present on dorsal surface; pronotum dorsally forming acute corner; mesoscutum humped, sparsely scattered with small punctures; lateral carina present on anterior mesoscutal surface; axilla and scutellum sparsely scattered with small punctures; metanotum setose; propodeum (Fig. 12) with pair of median, submedian and lateral carinae; median carina wider than other carinae, dorsal surface flattened (Fig. 12); median propodeal region narrower anteriorly, with several transverse carinae, posterior margin dorsally produced; submedian region with three transverse carinae, medially narrowed; lateral region with four transverse carinae.

Fore wing (Fig. 1) bearing four transverse dark bands; vein M 1.9 × basal part of RS; erect setae on C 0.2 × those on Sc+R and A.



Figures 1–3. *Ettchellsia ignita*, sp. n. (holotype). **1** General habitus **2** Head in dorsal view **3** Mesosoma in dorsal view. Scale = 1.0 mm.

Metasoma smooth but anterior surface of 6th metasomal tergite and 7–8th tergites entirely strongly shagreened; ovipositor 2.00 × mesosoma length, apex with small teeth and single knob.

Color. Head black; mandible black; antenna brown-black except scape and pedicel brownish; long setae on vertex and gena black, other setae white. Mesosoma black except brown tegula; long erect setae black; fore- and middle legs brown; hind leg with coxa, distal half of femur and distal four tarsomeres brown-black, trochanter and basal part of femur brown, tibia and basitarsus black; long setae on dorsal surface of hind tibia and basitarsus black. Metasoma and ovipositor reddish brown; ovipositor sheath pale brown.

Measurements. Head 1.05 mm long, 1.55 mm wide; mesosoma 2.10 mm long; scutum 1.35 mm wide; propodeal disc 0.70 mm long, 1.10 mm wide; fore wing 4.75 mm long; metasoma 2.30 mm long, 1.25 mm wide; ovipositor 4.20 mm long; total body length excluding ovipositor 5.45 mm.

(Male) Different from female as follows: Body brownish, head dark brown with brown antenna, mesosoma brown, legs testaceous excluding brown hind tarsus, metasoma testaceous; median carinae on propodeum narrower, dorsal surface rounded (as in Fig. 13).

Measurements. Head 0.76 mm long, 1.24 mm wide; mesosoma 1.75 mm long; scutum 1.10 mm wide; propodeal disc 0.55 mm long, 0.90 mm wide; fore wing 3.55 mm long; metasoma 1.40 mm long, 0.95 mm wide; total body length 3.77 mm.

Distribution. Peninsular Malaysia; Borneo.

Etymology. This species is named for the reddish coloration of the metasoma.

Remarks. This species is similar to *E. sinica* with both having a strongly humped mesoscutum, however, it is distinguished from the latter by the strongly narrowed median region of propodeum (Fig. 12); long erect black setae on hind tibia and basitarsus, many of which are longer than the width of the hind tibia (setae are whitish and shorter in *E. sinica*); reddish brown metasoma (Fig. 1) (metasoma is black in *E. sinica*).

Ettchellsia nigripes Mita & Shaw, sp. n.

urn:lsid:zoobank.org:act:1B9CC5DE-5AAC-483A-A952-B36A84C5DDFE\
http://species-id.net/wiki/Ettchellsia_nigripes
Figs 4–6, 13

Holotype ♀: "N. Sulawesi: Prov. Gorontalo, Pegunungan Tilongkabila, Bogani Nani Warta Bone N.P., 31. Jan-16 Feb 2010 (alt. 1200m), K. Takasuka leg. (Malaise trap)", "HOLOTYPE: *Ettchellsia nigripes* Mita & Shaw, 2012, sp. n.". The holotype is deposited in the Laboratory of Entomology, Faculty of Agriculture, Tokyo University of Agriculture, Atsugi, Japan.

Description. (Female) Head (Figs 4–5) 1.60 × wider than long, covered with long erect setae and short decumbent white setae; frons transversely rugose; surface around ocellar triangle smooth with rows of punctures behind anterior ocellus and outside of posterior ocelli; vertex reticulate-rugose; eye margined posteriorly by foveate groove and single post-ocular orbital carina; gena smooth with irregular carina under orbital carina; occipital carina not forming a depression; clypeus punctuate with small punctures.

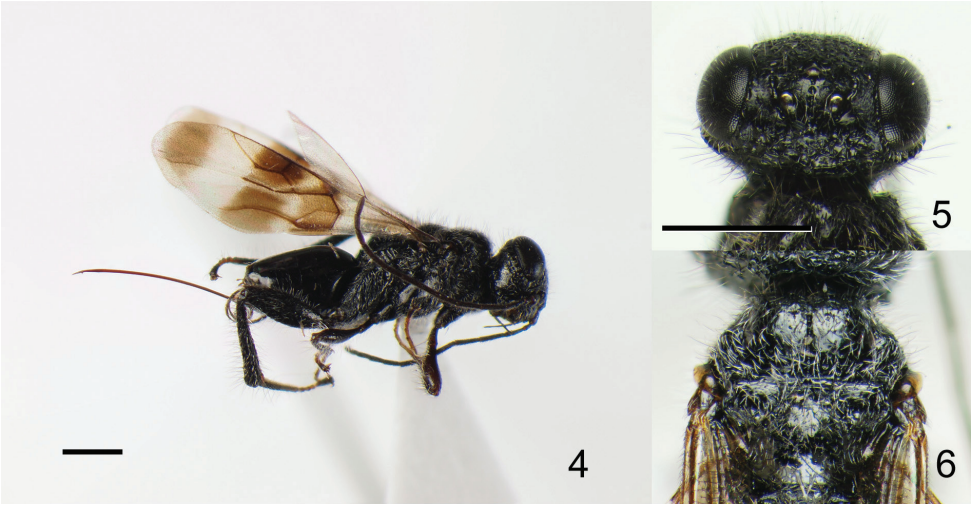
Mesosoma (Fig. 6) entirely covered with short decumbent setae, but long erect setae also present on mesonotum; mesoscutum smooth except lateral carina present on anterior surface; dorsal mesoscutal surface not humped; axilla and scutellum smooth; metanotum setose; propodeum (Figs. 13) with pair of median, submedian and lateral carinae; median region narrowed anteriorly, with eight indistinct transverse carinae, posterior margin posteriorly produced; submedian propodeal region converging posteriorly, with two transverse carinae excluding posterior areola; lateral region with two (right side) or four (left side) transverse carinae.

Fore wing (Fig. 4) bearing four transverse dark bands but separation between second and third bands indistinct; vein M 2.2 × basal part of RS; erect setae on C 1.2 × longer than those on Sc+R, equal to those on vein A.

Metasoma smooth except anterior surface of 6–8th metasomal tergites shagreened; ovipositor 1.29 × mesosoma length, apex with small teeth and single knob.

Color. Body entirely black except tarsi and ovipositor sheath dark brown, ovipositor reddish brown; long setae on vertex, gena, mesonotum and dorsal surface of hind tibia and basitarsus black, other setae white.

Measurements. Head 0.80 mm long, 1.55 mm wide; mesosoma 2.15 mm long; scutum 1.35 mm wide; propodeal disc 0.75 mm long, 1.10 mm wide; fore wing 4.85 mm long; metasoma 1.90 mm long, 1.20 mm wide; ovipositor 2.75 mm long; total body length excluding ovipositor 4.85 mm.



Figures 4–6. *Ettchellsia nigripes*, sp. n. (holotype). **4** General habitus **5** Head in dorsal view **6** Mesosoma in dorsal view. Scale = 1.0 mm.

(Male) Unknown.

Distribution. Sulawesi Island.

Etymology. This species is named for its black legs.

Remarks. This species is similar to *E. reidi* Mita & Shaw, sp. n. by the almost flat mesoscutum, however, it is distinguished from the latter by the anteriorly weakly narrowed median region of propodeum (Fig. 13) and the mostly smooth surface between posterior ocellus and eye (Fig. 5).

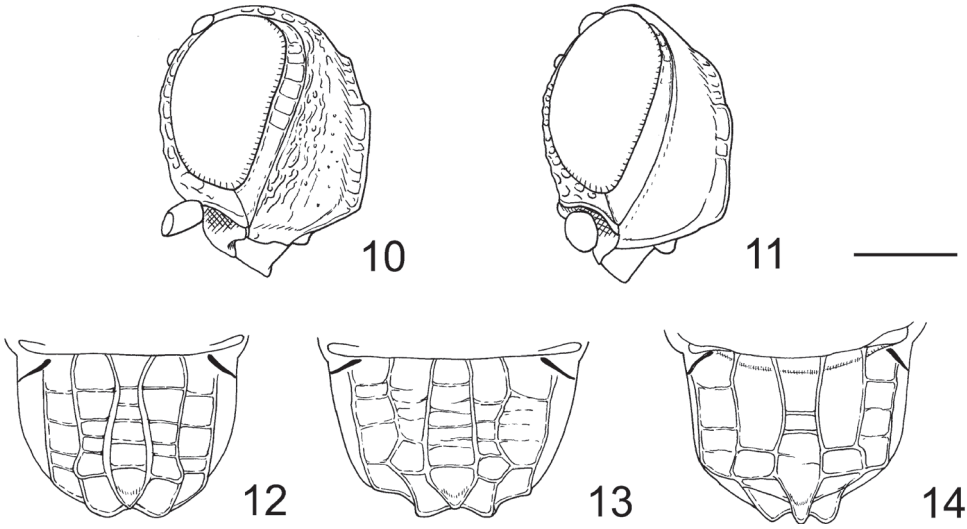
Ettchellsia reidi Mita & Shaw, sp. n.

urn:lsid:zoobank.org:act:403B45A6-E102-41E7-8A52-EBDC05C98BED
http://species-id.net/wiki/Ettchellsia_reidi
Figs 7–9, 14

Type series. Holotype ♀: "Sungai Sibau, nr entrance to Betung, Kerihun NP. ca. 4 km upstream, Kampung Putan. 21–27 Jun 1996, 70-90m. Chris Reid. IIS 967005", "20 yr. old lading, closed forest, Pan traps (yellow), 1°03'13"N, 113°00'56"E", "SEM", "HOLOTYPE: *Ettchellsia reidi* Mita & Shaw, 2012". Paratype: 1♀, same data as above, but Pan traps (yellow) 2:9, without label of "SEM". The holotype is deposited in the Museum Zoologi Bogor (MZB), the national museum of Indonesia. The paratype is deposited in the Royal Ontario Museum (ROM) Toronto, Canada.

Description. (Female) Head (Figs 7–8) 1.53–1.56 × wider than long, covered with long erect setae and short decumbent white setae; frons transversely rugose; surface around ocellar triangle rugose with rows of punctures behind anterior ocellus and outside of posterior ocelli, but lateral row indistinct among other sculptures; vertex

Figures 7–9. *Ettchellsia reidi*, sp. n. (holotype). **7** General habitus **8** Head in dorsal view **9** Mesosoma in dorsal view. Scale = 1.0 mm.



Figures 10–14. Heads and propodea of *Ettchellsia* spp. **10, 12** *E. ignita*, sp. n. **11** *E. piliceps* from Borneo **13** *E. nigripes*, sp. n. **14** *E. reidi*, sp. n. Scale = 0.5 mm.

reticulate rugose; eye margined posteriorly by foveate groove and single post-ocular orbital carina; gena smooth with irregular carinae under orbital carina; occipital carina dorso-laterally with a small depression; clypeus smooth.

Mesosoma (Fig. 9) entirely covered with short decumbent setae, but long erect setae also present on mesonotum; mesoscutum smooth; lateral carina present on anterior

surface; dorsal surface not swollen; axilla and scutellum smooth; metanotum setose; propodeum (Fig. 14) with pair of median, submedian and lateral carinae; median propodeal region narrower centrally, with a few transverse carinae in holotype, carinae indistinct in a paratype; posterior margin dorsally producing; submedian region smooth excluding posterior areola, parallel-sided; lateral region with at most four transverse carinae but keels sometimes indistinct.

Fore wing (Fig. 7) bearing three transverse dark bands with clear spot around Rs+M; vein M 1.6–1.7 × basal part of RS; erect setae on C 2.0 × those on Sc+R, 1.2 × those on A.

Metasoma smooth except anterior surface of 6–8th metasomal tergites shagreened; ovipositor 1.34–1.45 × mesosoma length, apex with small teeth and single knob.

Color. Head black; mandible dark brown; antenna dark brown except scape, pedicel and 5–8th flagellomeres brown; long setae on vertex, gena black. Mesosoma black; long setae on mesonotum black; legs black except tarsi brownish; long setae on dorsal surface of hind tibia and basitarsus black. Metasoma brownish black; ovipositor and ovipositor sheath dark brown.

Measurements. Head 0.80–1.10 mm long, 1.25–1.65 mm wide; mesosoma 1.55–2.20 mm long; scutum 0.95–1.30 mm wide; propodeal disc 0.65–0.85 mm long, 0.85–1.10 mm wide; fore wing 3.50–4.90 mm long; metasoma 1.55–2.35 mm long, 0.85–1.25 mm wide; ovipositor 2.25–2.95 mm long; total body length excluding ovipositor 3.75–5.45 mm.

(Male) Unknown.

Distribution. Kalimantan Barat, Borneo.

Etymology. The species name is dedicated to the collector of the types, Chris Reid, a coleopterist working at the Australian Museum, Sydney.

Remarks. The specimens were collected as part of the Insects of Indonesia Project, a collaboration of the ROM with the MZB. This species is similar to *E. nigripes* Mita & Shaw, sp. n. About identification of the two species, see the remarks of *E. nigripes*.

Acknowledgments

The authors express cordial thanks to Dr Keizo Takasuka and Mr Kyohei Watanabe from Kobe University, Kobe, Japan for providing the holotype of *Ettchellsia nigripes* sp. n. We are also grateful to Dr Christopher Darling (ROM), Dr David Smith (NMNH) and Mr. Takuma Yoshida (SEHU) for loaning the type and non-type material. Ms Natalie Dale-Skey Papilloud, Natural History Museum, London, UK loaned an identified female specimen of *E. piliceps* for comparison.

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ZooKeys 254: 109–120 (2012)
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Taxonomic study of the genus *Thisizima* Walker, 1864 in China, with descriptions of two new species (Lepidoptera, Tineidae)

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Academic editor: *E. van Nieukerken* | Received 5 September 2012 | Accepted 10 December 2012 | Published 21 December 2012

urn:lsid:zoobank.org:pub:0FEFD1A2-638F-4C50-A8D6-BCD07410E1AC

Citation: Yang L, Li H, Kendrick RC (2012) Taxonomic study of the genus *Thisizima* Walker, 1864 in China, with descriptions of two new species (Lepidoptera, Tineidae). ZooKeys 254: 109–120. doi: 10.3897/zookeys.254.3952

Abstract

The taxonomic study of the genus *Thisizima* Walker, 1864 is carried out in China. *Thisizima subceratella* **sp. n.** and *T. fasciaria* **sp. n.** are described as new based on the specimens collected in Fujian, Hainan and Hong Kong. Detailed male and female genitalia are described for the first time for the genus. Photographs of adults and genital structures are provided. A checklist of all the described species is included.

Keywords

Lepidoptera, Tineidae, *Thisizima*, new species, China

Introduction

The genus *Thisizima* was established by Walker (1864) with *T. ceratella* Walker, 1864 as the type species. It includes five named species: *T. ceratella* Walker, 1864 distributed in India, Burma, Thailand, West Malaysia and the Anambas Islands; *T. antiphanes*

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Meyrick, 1894 in Burma and Thailand; *T. sedilis* Meyrick, 1907 in Bhutan, Sikkim, Burma and Thailand; *T. bubalopa* Meyrick, 1911 in Sri Lanka and India, and *T. bovina* Meyrick, 1928 in the Andaman Islands (Robinson et al. 1994; Robinson 2008, 2009). Robinson (2009) further mentioned seven unnamed species occurring in India, the Andaman Islands, Thailand, Sarawak, Brunei and Hong Kong. Kendrick (2002) reported two *Thisizima* species occurring in Hong Kong in his PhD thesis: one was suspected to be *T. ceratella* Walker, 1864, another was unnamed. However, no detailed description of the genitalia has been given for the genus except that Robinson et al. (1994) mentioned that the female has an invaginated corethrogyne. We herein describe the generic characters in detail, add two new species, *T. subceratella* sp. n. and *T. fasciaria* sp. n. to the genus, and provide a checklist of *Thisizima* on a worldwide basis.

Material and methods

Specimens examined in this study were collected by light traps in Fujian, Hainan and Hong Kong. The type specimens are deposited in NKU and KFBG respectively.

Genitalia dissections were carried out following the methods described by Li (2002), and whole body dissections following methods described by Lee and Brown (2006). Photographs of the adults were taken with a Nikon D300 digital camera plus AF-S VR Micro-Nikkor 105mm f/2.8G IF-ED lens, and photographs of the genitalia were taken with an Olympus C7070WZ digital camera attached to an Olympus BX51 microscope.

Abbreviations

NKU Insect Collection, College of Life Sciences, Nankai University, Tianjin, China.
KFBG Kadoorie Farm and Botanic Garden, Hong Kong, China.
BMNH Natural History Museum, London, UK.
IMK Indian Museum, Kolkata (Calcutta), India.
HL Holotype.
PL Paratype.
ST Syntype.

Taxonomic accounts

***Thisizima* Walker, 1864**
http://species-id.net/wiki/Thisizima

Thisizima Walker, 1864: 820.

Type species: *Thisizima ceratella* Walker, 1864: 820, by monotypy.

Generic characters. Head with tufts of erect piliform scales. Antenna (Figs 5, 6) about 1.2× length of forewing in male, and 0.75× length in female; scape expanded, pecten with fewer than 15 bristles; flagellum compressed broadly and flatly, although considerably broader in male, with appressed scales, cilia not visible without removal of scales. Maxillary palpus rather reduced, minute, 1/3 length of first segment of labial palpus, 4-segmented, segmental ratio 2:1:1.5:1. Labial palpus (Figs 7, 8) rather long, almost 2.5× height of head, curved, ascending; segmental ratio 1:2:2; second segment with dense forwards projecting piliform scales, with 6–7 lateral bristles; third segment with appressed scales, slender and pointed, without vom Rath's organ. Forewing (Fig. 4) with costa moderately arched, apex roundly obtuse, termen obliquely rounded; unicolored or bicolored; all veins separate, R_1 from basal 1/4 of cell, R_3 from upper angle, R_5 to apex, M_3 close to CuA_1 at base, CuA_1 from lower angle of cell, forked portion of A_{1+2} about 1/3 length of vein, trace of CuP weak, cell closed, with trace of chorda and M stem; retinaculum in male subcostal, elongately triangular, with broad base and curled apex. Hind wing (Fig. 4) with costa moderately arched, apex rounded, slightly pointed than forewing, termen more oblique than forewing; all veins present, Rs parallel to $Sc+R_1$, CuA_2 from middle of lower margin of cell, trace of CuP weak, cell closed, with weak trace of M stem; frenulum with one stout bristle, sharp toward apex, angled at basal one third in male; also one bristle in female, much slender and shorter than in male. Legs smoothly scaled, except hind tibia with moderately elongate scales; foretibial epiphysis absent, hind tarsus without spine; tibial spur pattern 0-2-4, mid leg with outer spur about half length of inner spur, hind leg with outer mid spur about 0.4× length of inner spur, outer proximal spur about 0.6× length of inner spur.

Male genitalia. Corema absent in eighth segment. Vinculum rather broad, forming a strongly sclerotized cylinder with ill-defined tegumen. Uncus bilobate. Gnathos and subscaphium absent. Valva complex, base broad, scattered with strong hairs on outer surface; apodeme short, digitate. Transtilla absent.

Female genitalia. Corethrogyne present in seventh segment, invaginated into three intersegmental pouches. Ovipositor short and broad; papillae anales large, setose, sclerotized laterally; with one membranous dorsal sac and three ventral sacs between papillae anales, flower-like on top view. Apophyses short, apophysis posterior about 0.4× length of apophysis anterior. Ostium located on seventh sternite. Antrum distinct, anatropous.

Diagnosis. The elongate, curved, ascending labial palpus of *Thisizima* is quite similar to that of *Tinissa* Walker, 1864 in Scardiinae, to that of *Euplocamus* Latreille, 1809 and *Psecadioides* Butler, 1881 in Euplocaminae. *Thisizima* can be distinguished by the antenna with flagellum strongly flattened in both sexes; while the antenna is filiform in *Tinissa*, and bipectinate in male and filiform in female in Euplocaminae. Besides, R_4 and R_5 on the forewing are separated in *Thisizima*, but stalked in the last three genera. The corethrogyne of *Thisizima* in female is invaginated into three intersegmental pouches that can also be found in Perissomasticinae, but its other female characters are quite different from those of the members in Perissomasticinae.

Biology. Little is known about its biology. The living habitat is shown in figures 12 and 13.

Distribution. China (Fujian, Hainan, Hong Kong); Burma, Thailand, West Malaysia, India, Sikkim, Bhutan, Sri Lanka, the Andaman Islands and the Anambas Islands.

Remarks. *Thisizima* was placed in Tineidae since its establishment. The genus belongs to Tineidae without doubt, represented by the head with erect piliform scales, the subovate forewing with R_4 terminating on costa, the male retinaculum arising from Sc, and the female abdomen with corethrogyne in the seventh segment. However, its subfamily position has not been assigned due to some characters that indicate the uniqueness of this genus: the strongly flattened antenna, the rather reduced maxillary palpus, the absence of foretibial epiphysis and the position of ostium. The shape of the labial palpus may suggest its affiliation with Scardiinae and Euplocaminae; the corethrogyne in female may suggest its affiliation with Perissomasticinae. The status of *Thisizima* might be settled with further work on its morphology and biology.

Checklist of the genus *Thisizima* Walker, 1864

1. *Thisizima antiphanes* Meyrick, 1894
Thisizima antiphanes Meyrick, 1894: 27.
Type locality: Burma.
Depository of type: BMNH (HT).
Distribution: Burma and Thailand.

2. *Thisizima bovina* Meyrick, 1928
Thisizima bovina Meyrick, 1928: 428.
Type locality: Andaman Islands.
Depository of type: Unknown.
Distribution: Andaman Islands.

3. *Thisizima bubalopa* Meyrick, 1911
Thisizima bubalopa Meyrick, 1911: 125.
Type locality: Sri Lanka (Peradeniya); India (Nilgiris).
Depository of types: BMNH (ST).
Distribution: Sri Lanka and India.

4. *Thisizima ceratella* Walker, 1864
Thisizima ceratella Walker, 1864: 820.
Type locality: India.
Depository of type: BMNH (HT).
Distribution: India, Burma, Thailand, West Malaysia and Anambas Islands.

5. *Thisizima fasciaria* sp. n.
Type locality: China (Fujian).
Depository of type: NKU (HT & PT), KFBG (PT).
Distribution: China (Fujian, Hong Kong).

6. *Thisizima sedilis* Meyrick, 1907
Thisizima sedilis Meyrick, 1907: 989.
Type locality: Bhutan; Sikkim.
Depository of type: IMK (ST).
Distribution: Bhutan, Sikkim, Burma and Thailand.

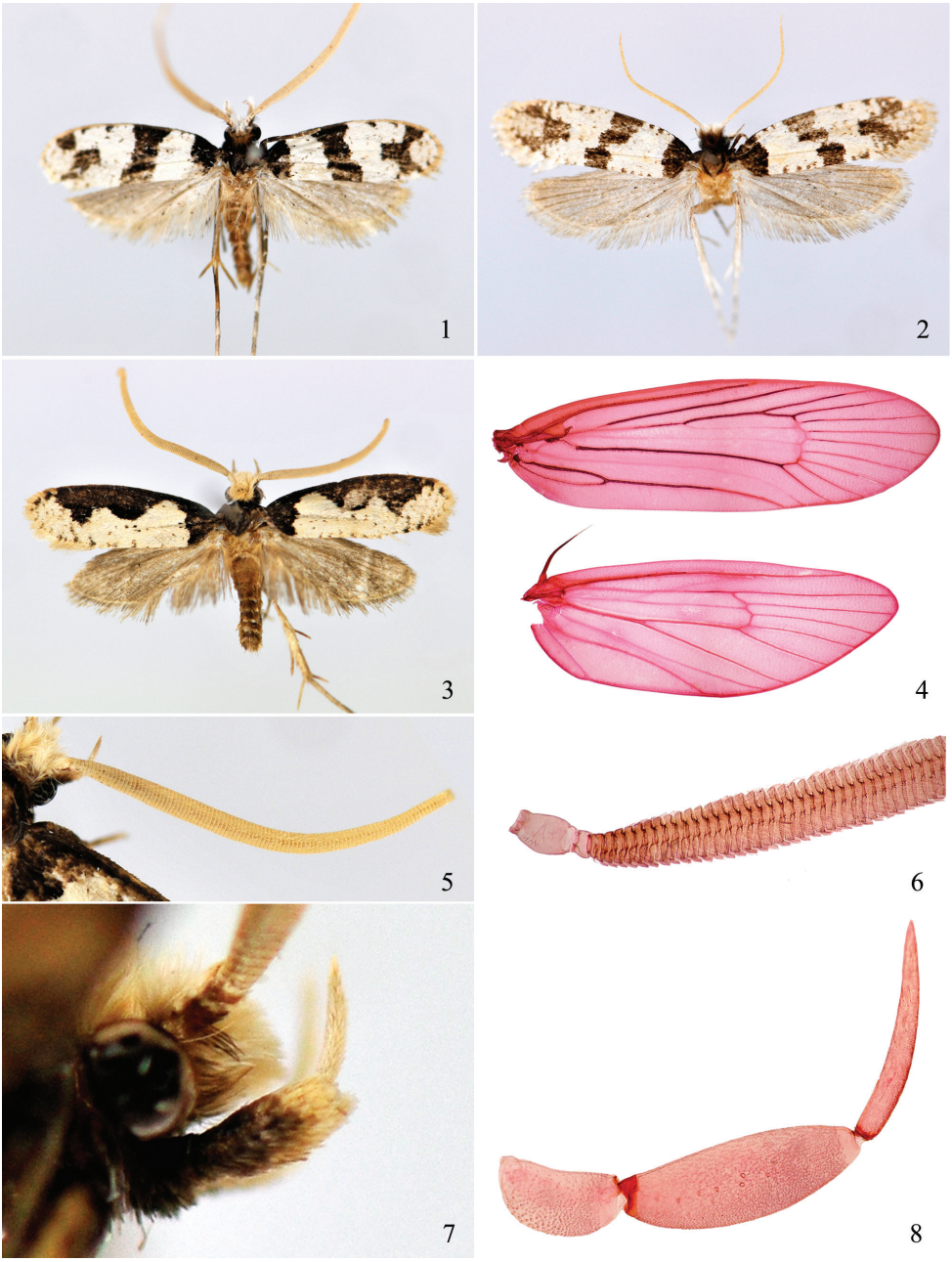
7. *Thisizima subceratella* sp. n.
Type locality: China (Fujian).
Depository of type: NKU (HT & PT), KFBG (PT).
Distribution: China (Fujian, Hainan, Hong Kong).

***Thisizima fasciaria* sp. n.**
urn:lsid:zoobank.org:act:A0755544-7CF8-4F5F-AE68-396A337FF41B
http://species-id.net/wiki/Thisizima_fasciaria
Figs 1, 2, 10–13

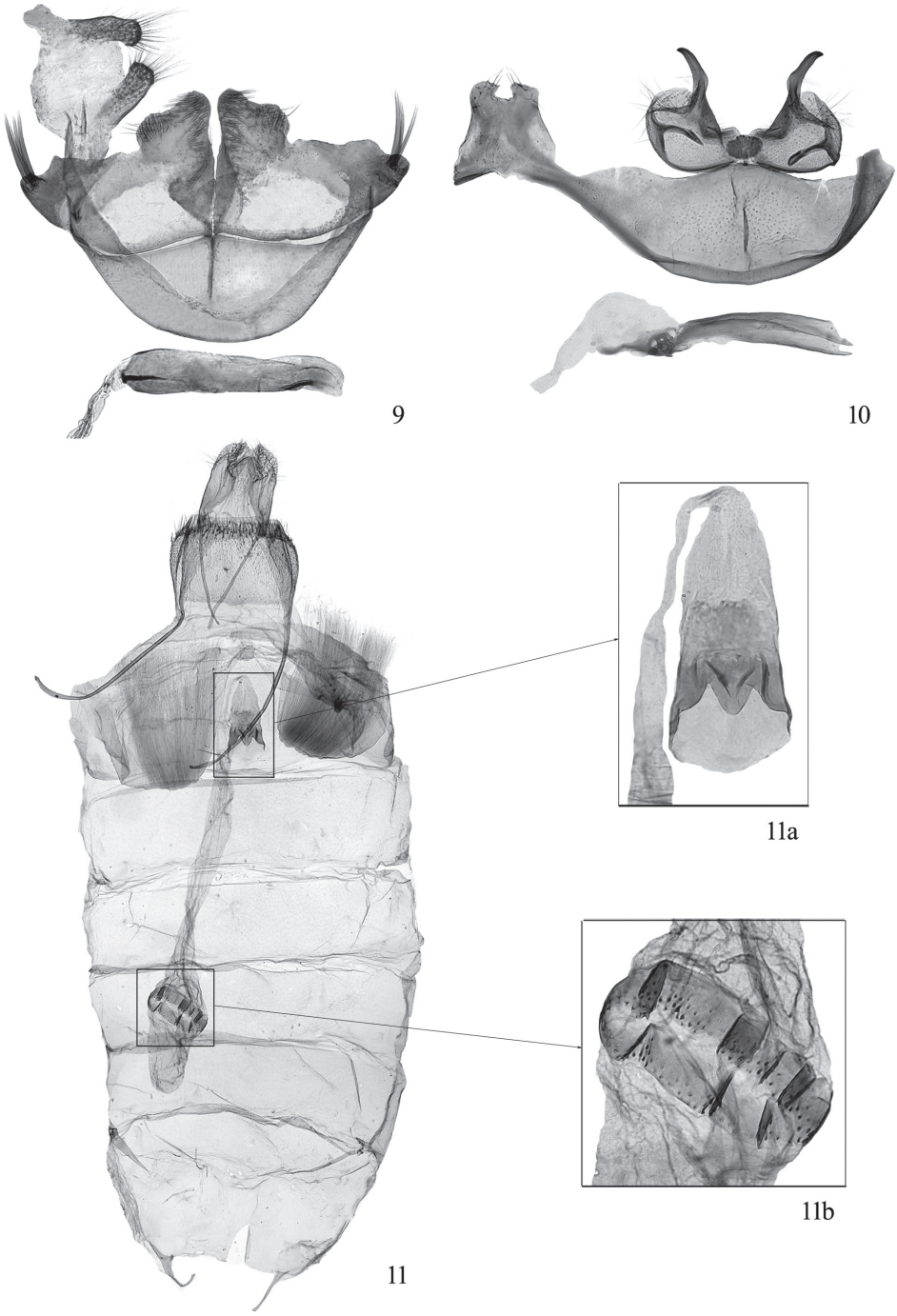
Type material. CHINA, Holotype ♂, **Fujian Province:** Mt. Tianzhu (24°35'N 117°55'E), Xiamen City, 220 m, 14.ix.2010, leg. Yinghui Sun & Jing Zhang (NKU).

Paratypes: 2 ♂, 15,19.viii.2010, leg. Bingbing Hu & Jing Zhang, same locality as holotype, genitalia slide No. YLL11172 (NKU). **Hong Kong:** 1 ♀, Kadoorie Agricultural Research Centre (22°25'N 114°06'E), 210 m, 20.iv.2007, leg. Houhun Li *et al.* (NKU), genitalia slide No. YLL11165; 1 ♂, Kadoorie Agricultural Research Centre, 210 m, 20.ix.2009, leg. Houhun Li *et al.* (NKU); 1 ♂, Kadoorie Farm and Botanic Garden (22°25'N 114°07'E), 315–575 m, 26.ix.2009, leg. Houhun Li *et al.* (NKU); 1 ♀, Kadoorie Agricultural Research Centre, Shek Kong, N.T., UTM: 50Q KV 030833, 28.iv.1997, 125W MBF, leg. R.C. Kendrick, genitalia slide No. YLL11171 (KFBG); 1 ♂, Kadoorie Agricultural Research Centre, Shek Kong, N.T., UTM: 50Q KK 029832, alt. 200 m, 6.v.1998, 125 W MBF, leg. R.C. Kendrick (KFBG); 1 ♂, Kadoorie Agricultural Research Centre, Shek Kong, N.T., UTM: 50Q KK 029832, alt. 200 m, 2.iv.1999, 125 W MBF, leg. R.C. Kendrick (KFBG).

Description. Imago (Figs 1, 2): Wingspan 13.0–15.0 mm in male, 17 mm in female. Vertex cupreous brown on posterior half, snow white on anterior half; frons snow white, with fine black scales on outer side before eyes. Antenna about 1.2× length of forewing in male, and 0.75× length in female including fringe; scape yellowish white above, brown mixed with black scales below, pecten with 10–15 black bristles; flagellum yellow, compressed broadly and flatly. Labial palpus snow white, first segment dark cupreous brown



Figures 1–8. Morphology of *Thisizima* spp. **1** *T. fasciaria* sp. n., holotype **2** *T. fasciaria* sp. n., paratype **3** *T. subceratella* sp. n., holotype **4–8** detailed structures of *T. subceratella* sp. n., paratypes: **4** vein **5, 6** antenna **7, 8** labial palpus (**4, 6, 8** slide No. NKYLL010) (**1, 3–8** ♂, **2** ♀).



Figures 9–11. Genitalia of *Thisizima* spp. **9** male genitalia of *T. subceratella* sp. n., paratype, slide No. NKYLL010 **10–11** *T. fasciaria* sp. n., paratypes: **10** male genitalia, slide No. YLL11172 **11** female genitalia (11a. antrum; 11b. signum), slide No. YLL11156.

on outer surface, second segment cupreous brown on outer surface of basal half, with sparse black lateral bristles. Thorax and tegula black. Forewing index about 0.32; ground color bright white; a black triangular patch from costal margin to dorsum on basal 1/6; an oblique, black fascia from basal 1/3 to just before middle of dorsum, slightly narrowed medially, sinuate along both margins; a rectangular black patch from outer margin of cell to distal 1/6 of forewing, confluent with two black subtriangular patches from costa and termen before apex respectively, forming a broad Y-shaped pattern; two black costal spots between oblique fascia and Y-shaped pattern; termen and dorsum scattered with faint dark brown dots, dim in some specimens; fringe yellowish brown. Hindwing index 0.35; light grayish brown; fringe gray; frenulum with one stout bristle in male, one much slender and shorter bristle in female. Fore leg black; mid leg black, with snow white fine scales at apex of tibia, tarsus yellowish brown on ventral surface, with white at apex of each segment on dorsal surface, spurs dark brown; hind leg and spurs yellowish brown, apex of tibia and each segment of tarsus with white scales dorsally.

Male genitalia (Fig. 10). Vinculum convex anteriorly in arch, slightly convex at middle on posterior margin. Uncus sclerotized, trapezoidal, semicircularly concave at middle on posterior margin; uncus lobes short, setose. Valva rounded in basal half, horn-shaped and curved outwardly in distal half, setose on outer surface. Juxta small, subovate. Aedeagus slightly curved ventrad, with a pair of blade-shaped lateral sclerites connected by membrane dorsally and opened ventrally, apex sharp obliquely; cornutus absent.

Female genitalia (Fig. 11). Seventh tergite short, membranous except heavily sclerotized anteriorly; sternite slightly sclerotized, 0.2× as long as broad, tapered posteriorly, produced to a slender, elongate plate at middle. Eighth tergite rectangular, with dense, minute spinules, with short spines along posterior margin; sternite rectangular, with dome-like membranous section medially, scattered with short spinules, sclerotized section with dense short spinules. Ostium at anterior 1/3 of seventh sternite, rounded. Antrum (Fig. 11a) heavily sclerotized, funnel-shaped, with a more sclerotized pouch anteriorly on ventral surface. Ductus bursae about 1.4 × length of corpus bursae, posterior 1/6 slender, anterior 5/6 broadened; inception of ductus seminalis at posterior 1/6 of ductus bursae. Corpus bursae elongate oval, with a broad ring-shaped, punctate signum at posterior 1/3 (Fig. 11b).

Diagnosis. The bold, bicolored forewing pattern of the new species is diagnostic: ground color bright white with a triangular black patch at base, an oblique, black fascia near middle and a somewhat Y-shaped black patch near apex. There are four other species with bicolored forewing pattern: *T. antiphanes* has a white basal band and an apical dot, *T. sedilis* has a large rounded-triangular costal blotch, *T. ceratella* has a broad irregular costal stripe broadly confluent with basal patch on anterior margin, and *T. subceratella* sp. n. has a broad black costal blotch narrowly confluent with basal patch on anterior margin.

Biology. The living habitat is shown in figures 12 and 13.

Distribution. China (Fujian, Hong Kong).

Etymology. This specific name is derived from the Latin *fasciarius*, meaning fascia, referring to the oblique, black fascia near middle of forewing.

Figures 12–13. Adults of *Thisizima fasciaria* sp. n. in repose. **12** Shan Liu, Sai Kung, Hong Kong, 9.iv.2010, photographed by R.C. Kendrick **13** Hong Kong, photographed by R.C. Kendrick.

***Thisizima subceratella* sp. n.**
urn:lsid:zoobank.org:act:FA0F81DD-9A18-4261-BB92-F89F9681B288
http://species-id.net/wiki/Thisizima_subceratella
Figs 3–9

Type material. CHINA, Holotype ♂, **Fujian Province**: Mt. Tianzhu (24°35'N 117°55'E), Xiamen City, 220 m, 12.ix.2010, leg. Yinghui Sun & Jing Zhang (NKU).

Paratypes: 2 ♂, 30.viii,19.ix.2010, other same data as holotype, genitalia slide No. NKYLL010 (NKU). **Hainan Province:** 1 ♂, Mt. Wuzhi (18°31'E, 109°24'E), 700 m, 19.v.2007, leg. Zhiwei Zhang & Weichun Li (NKU). **Hong Kong:** 2 ♂, Kadoorie Agricultural Research Centre (22°25'N 114°06'E), Shek Kong, N.T., UTM: 50Q KV 030833, 125 W MBF, 28.iv.1997, leg. R.C. Kendrick (KFBG).

Description. Imago (Figs 3–8). Wingspan 18.5–20.5 mm in male. Vertex and frons yellowish white, tinged with brown around eyes. Antenna about 1.2× length of forewing including fringe; scape yellowish white above, scattered with brown scales below, pecten with 8–12 black bristles; flagellum ochreous yellow, compressed broadly and flatly. Labial palpus yellowish white, first segment black on outer surface, second segment black on outer surface of basal 2/3, with sparse black lateral bristles. Thorax black, tegula black on anterior half and dark cupreous brown on posterior half. Forewing index about 0.3; ground color creamy white, with scattered dark brown scales on distal 1/4; a triangular black patch on basal 1/5, broader at costa, sinuate along outer margin; a broad black costal blotch narrowly confluent with basal patch on anterior margin, curved in W shape on posterior margin, its basal half apically rounded and reaching half width, distal half triangularly crossing midwing; two small indistinct spots along costa before apex; termen and dorsum scattered with faint dark brown dots; fringe light yellowish brown. Hind wing index 0.38; cupreous brown; fringe grayish brown; frenulum one stout bristle. Fore leg black except distal half of coxa yellowish brown on ventral surface; mid leg black except tarsus yellowish brown on ventral surface, spurs dark brown; hind leg yellowish brown mixed with black scales.

Male genitalia (Fig. 9). Vinculum convex anteriorly in arch, slightly convex at middle on posterior margin; with a longitudinal suture in middle posteriorly, 0.5× length of vinculum. Uncus membranous, trapezoidal; uncus lobes digitate, slightly sclerotized, with elongate setae on ventral surface. Valva with base broad and gently arched, connected ventrally at base by a sclerotized, somewhat funnel-shaped plate on inner surface; basal 2/3 membranous and inverted fan-shaped, scattered with strong hairs on outer surface; apex as wide as base, heavily sclerotized, sinuate, dorsoapically with a tuft of 3–4 strong spines, ventroapically convex, with dense, stout spines on outer surface. Juxta membranous. Aedeagus membranous dorsally, sclerotized ventrally, with a shallow keel at base; cornutus absent.

Female. Unknown.

Diagnosis. The new species is quite similar to *T. ceratella*, but can be separated by the forewing having a broad blackish costal blotch narrowly confluent with the basal patch on anterior margin, curved in W-shape on posterior margin; while *T. ceratella*

has an irregular cupreous black costal stripe broadly confluent with the basal patch on anterior margin, curved outward before middle on posterior margin.

Distribution. China (Fujian, Hainan, Hong Kong).

Etymology. The specific name is derived from the Latin prefix *sub-*, meaning similar, and another specific name *ceratella*, referring to the similarities of the two species.

Remarks. K. Tuck (BMNH) assisted us to check the identity of *T. ceratella*. Unfortunately, the holotype has lost its hindwings and abdomen. The late G. Robinson had therefore dissected a male specimen identified as *ceratella* in the Meyrick collection, collected in Koni, Burma. Tuck kindly compared our illustrations of *T. subceratella* sp. n. with Robinson's slide BMNH Microlep. No. 27736. He noticed a small but distinct difference in the shape of the valva: in Robinson's dissection the valva is slightly larger and therefore extends further laterally and has five strong spines, whereas our illustration shows a relatively short valva with only 3–4 spines on each.

Furthermore, the adult photograph of *T. ceratella* given by Robinson *et al.* (1994) shows that the forewing pattern is coincident with Walker's original description. We thus base our understanding of the identity of *T. ceratella* on this dissected specimen.

There are many tineid species showing small differences in genitalia, but they can usually be recognized by the external morphology, such as forewing pattern and venation (eg. species of *Monopis* Hübner). The forewing pattern in the new species is quite different from that of *T. ceratella*, and the shape of the valva does have small but distinct difference between the two species, which we regard as sufficient evidence that this is a good species.

Acknowledgements

We express our thanks to Mr. Kevin Tuck (BMNH, London) for checking the holotype of *Thisizima ceratella*, to anonymous referees for their helpful comments and precious references, and to those who took part in the field work. This research was supported by the National Natural Science Foundation of China (No. 30930014 & No. J1210005).

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