

A review of the genus *Metalype* Klapálek, with descriptions of three new species from China (Trichoptera, Psychomyiidae)

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Abstract

Three new species of *Metalype* from China, *Metalype hubeiensis* Qiu & Morse, **sp. n.**, *M. shexianensis* Qiu & Morse, **sp. n.**, and *M. truncata* Qiu & Morse, **sp. n.**, are described and illustrated. *Metalype uncatissima* (Botosaneanu, 1970) is reported from China for the first time. The differences between genus *Metalype* and genus *Psychomyia* are discussed and four *Psychomyia* species are transferred to *Metalype*: *Metalype bolzenthali* (Schmid, 1997); *M. klapaleki* (Malicky, 1995a); *M. kumari* (Schmid, 1997); and *M. nithaiaha* (Malicky, 2014). A key to the males of *Metalype* species of the world is provided.

Keywords

Annulipalpia, caddisfly, east Palearctic Region, Oriental Region

Introduction

Knowledge of the Chinese Trichoptera fauna was limited before the mid-1900s, described solely by foreign scholars (Morse et al. 1994). It has increased considerably since the 1980s, mostly due to the work of Chinese scientists. There were only 530 Chinese

species known by 1990 (Yang et al. 2005), but 1267 Chinese species were described by the middle of 2014 (Yang et al. 2016). However, records of Psychomyiidae increased from 19 species to only 26 species in that interval; this number is relatively small compared to the number of Psychomyiidae species known from the Oriental and East Palearctic Regions (405 spp., Morse unpublished data) and from adjacent countries (e.g., 73 spp. in India, 58 in Thailand, 35 in Vietnam; Morse unpublished data). Schmid (1984) estimated that there are actually 40,000 caddisfly species in southwestern Asia, although this estimate has been questioned by Malicky (1993a). Thus, this study is part of a continuing effort to document the Chinese caddisfly fauna that is mostly unknown to science, focusing here on *Metalype* of Psychomyiidae.

The genus *Metalype* was established by Klapálek (1898). For more than 100 years, it contained only the type species *Metalype fragilis* (Pictet, 1834). Wing venation (Fig. 1) and male genitalia of *Metalype* are very similar to those of *Psychomyia* Latreille, 1829 (in Cuvier 1829; type species *Psychomyia annulicornis* Pictet, 1834, selected by Ross 1944, synonym of *Psychomyia pusilla* Fabricius, 1781). Malicky (1995a) suggested that *Metalype* is a synonym of *Psychomyia*. Schmid (1997) treated *M. fragilis* as a *Psychomyia* species and included it in his *Psychomyia mahayinna* species group (“*Mahayinna* Group”) with six other *Psychomyia* species; he suggested that this group is the oldest lineage of *Psychomyia*. Li and Morse (1997) completed a phylogenetic analysis of Psychomyiidae and concluded that *Metalype* is a monophyletic genus closely related to *Psychomyia* and *Paduniella* Ulmer, 1913 (type species *Paduniella semarangensis* Ulmer, 1913, monotypic), these three genera collectively constituting the subfamily Psychomyiinae. Li and Morse (1997) also listed characters supporting the monophyly of *Metalype* and transferred three *Psychomyia* species to *Metalype*. Later, they indicated that *Metalype* and *Psychomyia* are sister genera, and *Metalype* + *Psychomyia* is the sister lineage to *Paduniella* (Li and Morse 1998). However, some *Metalype* species are still considered to belong in *Psychomyia* by some authors (Robert 2002, Mey and Nozaki 2006, Waringer and Graf 2011, Malicky 2014). Frandsen et al. (2016) concluded that Psychomyiinae is monophyletic, but in addition, they included the genus *Lype* McLachlan, 1878 (type species *Lype phaeopa* (Stephens, 1836), selected by Ross 1944) as sister to *Paduniella* in their phylogeny of this subfamily.

In Asia, *Metalype* species have been reported from Japan (Mey and Nozaki 2006, Nozaki and Nakamura 2007), Korea (Botosaneanu 1970), Nepal (Malicky 1995b), Pakistan (Schmid 1961), and Russia (Levanidova et al. 1995), but not from China (Yang et al. 2016); this apparent absence may have resulted from a lack of studies, or *Metalype* species are recognized in China as species of *Psychomyia*. For example, *Psychomyia nithaiiah* Malicky, 2014 was described from Taiwan, but it is probably a *Metalype* species because it is very similar to *Metalype uncatissima* (Botosaneanu, 1970). In this article, we report four *Metalype* species from China, with three of them new to science. We also discuss the differences between *Metalype* and *Psychomyia* species. A key to males of *Metalype* species of the world is also provided.

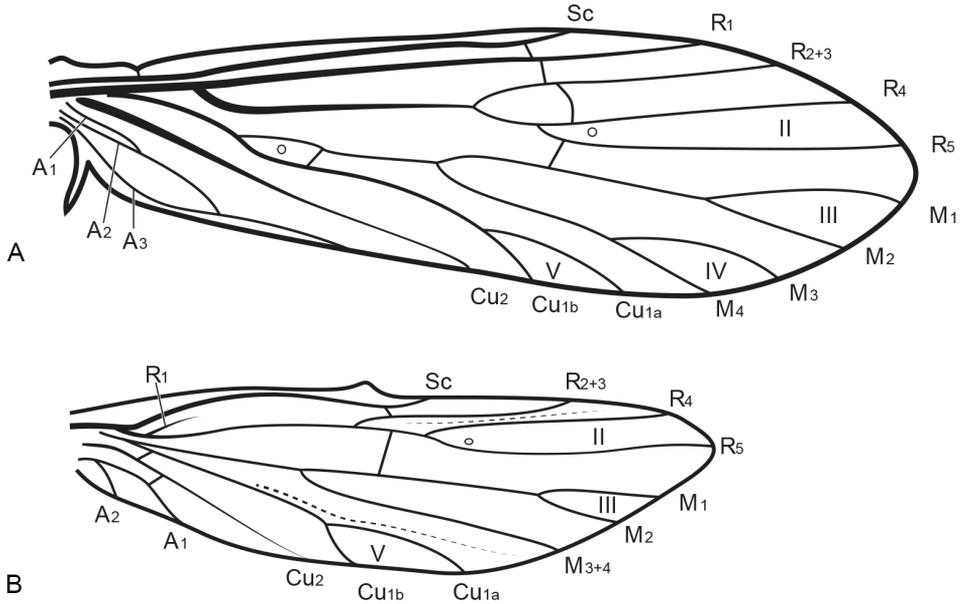


Figure 1. Wing venation of *Metalype truncata* sp. n., right wings, dorsal. **A** forewing **B** hind wing.

Methods

The three new species were first described in Dr Li You-wen's dissertation (Li 1998), but their names were explicitly excluded from availability under Article 8 of the 3rd edition of the International Code of Zoological Nomenclature (International Commission on Zoological Nomenclature 1985). However, Dr Li deposited his material in the Clemson University Arthropod Collection (CUAC), Clemson, South Carolina, USA; and Department of Plant Protection, Nan-jing Agriculture University (NJAU), Nan-jing, People's Republic of China (PRC). Here these species are described based on those specimens to make the names available.

Specimens were collected with ultraviolet light traps during 1990–1993 and were preserved in 80% ethanol. The sampling sites are listed in Table 1, with original label names and modern or corrected Chinese names. Holotypes of the new species are deposited in NJAU, paratypes are deposited in NJAU and CUAC. The specimens of *Psychomyia klapaleki* Malicky, 1995a and *Metalype fragilis* were loaned by Dr Hans Malicky from his personal collection in Lunz am See, Austria.

Specimens are all preserved in 75%–100% ethanol. Abdomens of a few individuals were removed and water-bath heated in 10% KOH for a few minutes to remove muscle and other non-chitinous tissues for illustration. Specimens were observed under a dissecting microscope. An eyepiece with a grid was used to prepare pencil templates of the various views. The templates were traced with the vector graphics software Adobe Illustrator® (version 19.0.0, 64-bit).

Table 1. Locations of *Metatype* species from China.

Species	Province	County	[Geographic coordinate]	Notes	Elevation
<i>Metatype hubeiensis</i>	Hu-bei Province 湖北省	[Jing-shan-xian] (Jin-shan-xian) 京山县	[31°16.74'N; 113°12.20'E]	[San-yang Town], Da-fu-shui [三阳镇,] 大富水	90 m
<i>Metatype shexianensis</i>	An-hui Province 安徽省	She-xian 歙县	[30°1.19'N; 118°17.84'E]	Yang-jia-tan, Feng-yuan-shui 杨家坦, 丰源水	215 m
			[30°5.94'N; 118°21.54'E]	Yan-yuan Town, Huang-bai-shan Village 岩源镇, 黄柏山村	[717 m]
<i>Metatype truncata</i>	Si-chuan Province 四川省	[Jiu-zhai-gou-xian] (Nan-ping-xian) [九寨沟县] (南坪县)	[33°16.02'N; 103°55.08'E]	Jiu-zhai-gou [National Park] 九寨沟[国家公园]	2000 m
		[Du-jiang-yan-shi] (Guan-xian) [都江堰市] (灌县)	[30°53.90'N; 103°34.37'E]	Qing-cheng-shan Town, Wei-jiang-he 青城山镇, 味江河	930 m
<i>Metatype uncatissima</i>	Hei-long-jiang Province 黑龙江省	Shang-zhi-xian 尚志县	[45°16.40'N; 127°30.26'E]	Mao-er-shan Town, A-shi River 帽儿山镇, 阿什河	300 m
			[44°39.33'N; 128°13.90'E]	Wei-he Town, Yu-lin Tree Farm 苇河镇, 榆林林场	380 m
		Tie-li-shi 铁力市	[46°37.58'N; 129°7.29'E]	Lang-xiang Town, Ba-lan Farm 朗乡镇, 巴兰农场	160 m
		Yi-chun-shi 伊春市	[48°37.09'N; 129°32.96'E]	Wu-yi-ling, Wu-yun River 乌伊岭, 乌云河	160 m

[] = information that was not written on the original labels, including modern name or correctly spelled name; () = abandoned name, or name wrongly spelled on the original labels.

For the specimens that were collected during 1990–1993, no geographical coordinates were taken by GPS at that time. We tried to find the most probable sampling sites based on the location names and descriptions of original labels, and obtained the geographical coordinates from Google Earth (Version 7.1.7.2600). The elevation of one site: An-hui Province, She County, Yan-yuan Town, Huang-bai-shan Village, was missing, so the elevation of this site was also obtained from Google Earth. Elevations of all other sites were obtained from the labels. Modern Chinese names and geographical coordinates of sampling sites were confirmed by Prof Sun Chang-hai (Sun C-h) of Nan-jing Agriculture University.

Terminology for wing venation (Fig. 1) follows that of Schmid (1998). Terminology for male genitalia follows Nielson (1957) except that the pair of flat processes beyond the superior appendages are called “Tergites IX+X” (Ross 1938) and the apical portion of the phallus is called a “phallicata” (“phallicata” [sic], Ross 1956). Terminology for larvae follows Wiggins (1996).

Results

Metalype hubeiensis Qiu & Morse, sp. n.

<http://zoobank.org/5320A02D-CC87-4ACE-BD2F-0A48DDCBCBD9>

Fig. 2A–E

Metalype hubeiensis Li, 1998: 223–224, figs 11.21–11.24, *nomen nudum*.

Type locality. PRC, Hu-bei Province: Jing-shan County, tributary of Da-fu-shui River, 50 km NW of Ying-cheng downtown, 31°16.74'N; 113°12.20'E, 90 m, 17 July 1990, collector JC Morse.

Type specimen. Holotype. Male, in 75% ethanol, in cotton-stoppered microvial inside screwcap vial. Original label: “Hú běi, Jīn-shān-xiàn, 50 KM N.W. of Yīn-chéng, Trib. of Dà-fù-shǔi, 17 July 1990, 90 M elev., coll. Morse” “*Metalype hubeiensis* Holotype Li & Morse”. Deposited in NJAU.

Paratypes. Same data as holotype, 5 males (4 in CUAC, 1 in NJAU). Original label: “Hú běi, Jīn-shān-xiàn, 50 KM N.W. of Yīn-chéng, Trib. of Dà-fù-shǔi, 17 July 1990, 90 M elev., coll. Morse” “Psychomyia sp. 7 鉴定者” [genus and species identity handwritten, Chinese characters = “Identifier”] “*Metalype hubeiensis* sp. n. paratype Li & Morse” [author names handwritten]. Also, a red paper tag without writing. Deposited in NJAU.

Diagnosis. This species resembles *Metalype truncata* sp. n. The differences are as follows: (1) The apicomesal spur on each hind leg of *M. hubeiensis* is curved mesad and forked apically (Fig. 2E; the apicomesal spur on each hind leg of *M. truncata* is truncate apically, with a few lobes and an acute process, Fig. 4E); (2) in ventral view the coxopodites of *M. hubeiensis* are fused with each other basally (Fig. 2D; in ventral view the coxopodites are fused with each other for more than half of their length in *M. truncata*, Fig. 4D); (3) in lateral view the harpagones of *M. hubeiensis* are slightly expanded in the middle dorsally, each less than two times as wide as the basal part (Fig. 2A; in lateral view the harpagones of *M. truncata* are strongly expanded in the middle dorsally, each more than two times as wide as the basal part, Fig. 4A); and (4) in ventral view the harpagones of *M. hubeiensis* are hooked mesodorsad (Fig. 2D; in ventral view the harpagones of *M. truncata* are hooked mesad, Fig. 4D).

Description. Male. Forewings each 3.4–3.9 mm (n = 5). Compound eyes black, body yellow. Apicomesal spur of each hind leg slightly curved mesad and forked apically.

Genitalia. In lateral view tergites IX+X wide basally, in dorsal view each half triangular and slightly narrowed laterally at two-thirds distance from base. In lateral view superior appendages digitate, wide basally and gradually narrowed from base to apex; in dorsal view central part slightly concave laterally, setose and with few stout and curved setae at apex; each with subapicomesal tooth short, about as long as wide. In ventral view sternite IX slightly expanded posteriorly. In lateral view coxopodites triangular, in ventral view subrectangular and fused with each other only basally. In

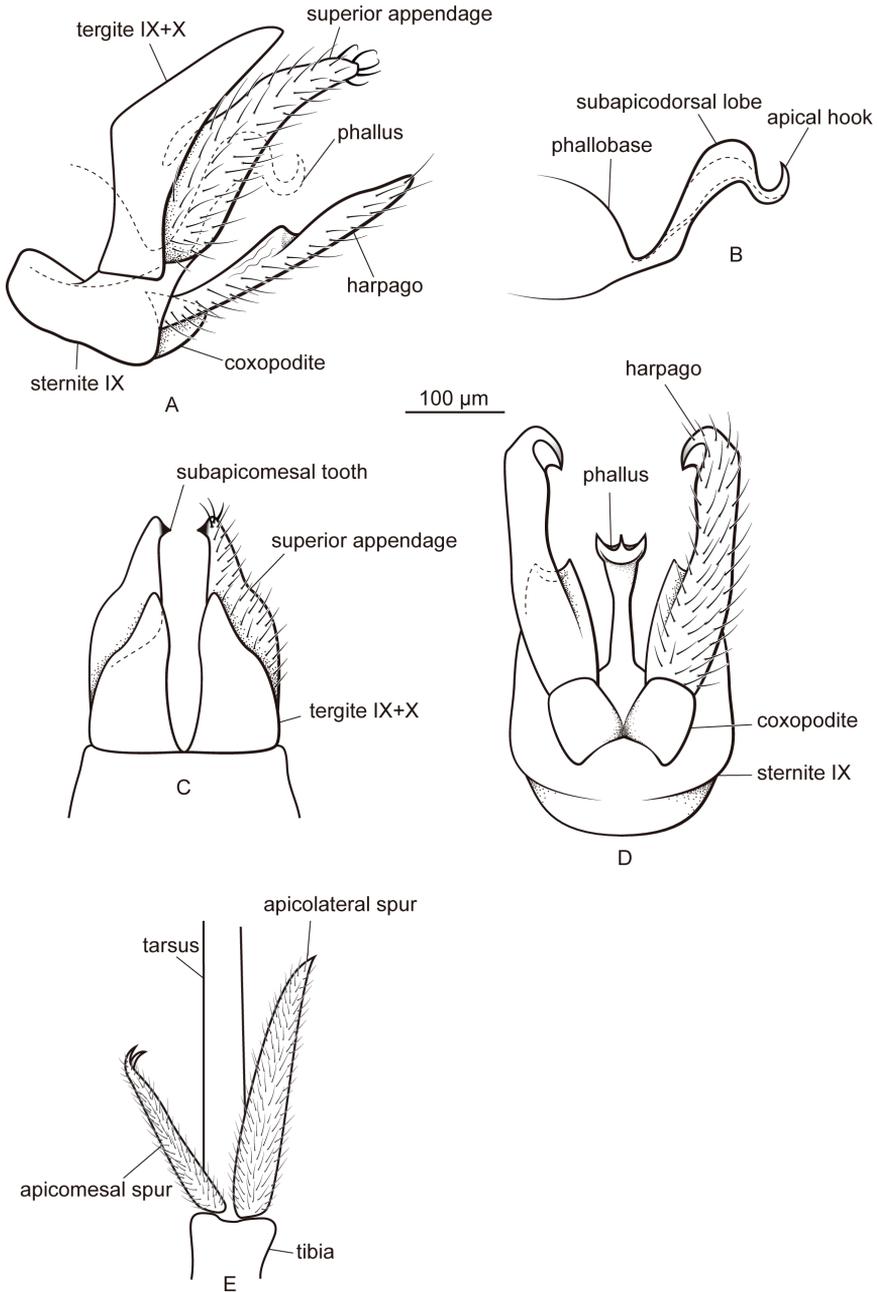


Figure 2. Male of *Metatypa hubeiensis* sp. n. **A** genitalia, left lateral **B** phallus, left lateral **C** genitalia, dorsal **D** genitalia, ventral **E** apical spurs of right hind leg, ventral.

lateral and ventral views, harpagones each weakly sclerotized and slightly expanded mesodorsad at mid length, in ventral view slightly curved mesad and strongly hooked mesad apically, with harpagonal hook stout and its mesal edge membranous, slightly sclerotized at apex. In lateral view phallus with two major curves, both curves greater than 90°, phallobase expanded, phallicata with pair of round subapicodorsal lobes and apical hook directed dorsad.

Female. Unknown.

Etymology. An adjective in nominative singular from “Hu-bei,” a province in China, referring to the type locality of this species.

Distribution. This species has been found only at the type locality, Jing-shan County, Hu-bei Province, southcentral China, Oriental Region.

***Metatype shexianensis* Qiu & Morse, sp. n.**

<http://zoobank.org/D13F26EB-54B7-41B2-AB94-AD067997D3ED>

Fig. 3A–F

Metatype shexianensis Li, 1998: 221–222, figs 11.13–11.16, *nomen nudum*.

Type locality. PRC, An-hui Province: She County, Yang-jia-tan, Feng-yuan-shui stream, 30°1.19'N; 118°17.84'E, 215 m, 24 May 1992, Collector JC Morse and Sun C-h; She County, Yan-yuan town, Huang-bai-shan village, Feng-yuan-shui stream, 30°5.94'N; 118°21.54'E, 717 m, 14 June 1991, collector Li Y-w.

Type specimen. Holotype. Male, in 75% ethanol; head and prothorax, wings, cleared genitalia in different cotton-stoppered microvials inside one screwcap vial. Original label: “Ānhūi Shè-xiàn, Yáng-jīā-tán, Fēng-yuán-shuǐ, 215 M elev., 24 May, 1992, Coll. Morse, Sun” “*Metatype shexianensis*, Holotype, Morse & Sun 1992”. Deposited in NJAU.

Paratypes. 2 males, in 80% ethanol, in cotton-stoppered microvial inside screwcap vial; one specimen cleared. Original label: “晚歙县岩源, 黄柏山村, 1991. 6-14” [Handwritten, Chinese characters = “Night She County Yan-yuan, Huang-bai-shan Village”] “*Metatype shexianensis* sp. n., Paratypes, Li & Morse 1996”. Deposited in CUAC.

Diagnosis. This species resembles *Metatype anaktujuh* (Malicky, 1995b) (Malicky 1995b, page 23, figures in the top right corner) but can be distinguished by the following characters: (1) In lateral view the harpagones of *M. shexianensis* are slightly narrower in the middle than at the ends (Fig. 3A) in contrast to the harpagones of *M. anaktujuh* (Malicky 1995b, page 23, figure on the left); (2) In dorsal view the harpagones of *M. shexianensis* each bears two mesal processes, the anterior one is larger and truncate, the posterior one smaller and digitate (Fig. 3C), whereas the harpagones of *M. anaktujuh* each bears one truncate mesal process (Malicky 1995b, page 23, figure on the right).

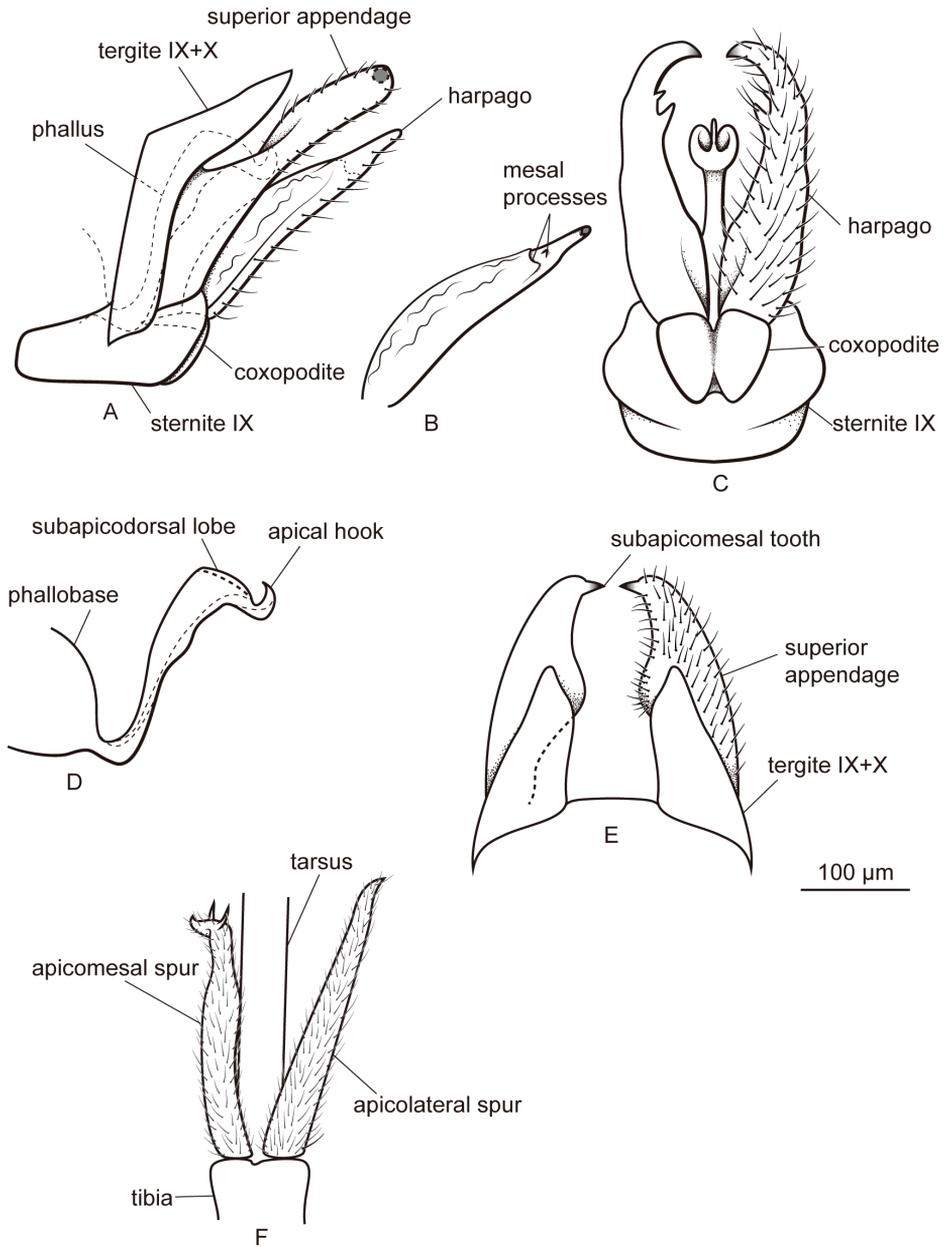


Figure 3. Male of *Metatype shexianensis* sp. n. **A** genitalia, left lateral **B** right harpago, mesal **C** genitalia, ventral **D** phallus, left lateral **E** genitalia, dorsal **F** apical spurs of right hind leg, ventral.

Description. Male. Forewings each 3.8–3.9 mm ($n = 2$). Compound eyes black, body yellow. Apicomesal spur of each hind tibia curved laterad and twisted apically, with two small subapical processes.

Genitalia. In dorsal view tergites IX+X widely separated from each other, each half triangular, in lateral view nearly L-shaped. In lateral view superior appendages setose, each wide at base, narrower at mid length than at the ends and digitate at apical half; in dorsal view mid length expanded mesally and covered with short setae; subapicomeral teeth each about two times as long as wide. In ventral view sternite IX slightly expanded posteriorly. In lateral view coxopodites triangular, in ventral view ovate and fused with each other for over half of their length. In lateral view harpagones slightly shorter than superior appendages, weakly sclerotized dorsally and tapered to apex, setose ventrally; in ventral view slightly expanded basomesally, curved mesad and slightly sclerotized at apices, each with two mesal processes subapically, anterior one larger; in mesal view truncate with notch, posterior one small, digitate, bearing few setae at apex. Phallobase expanded, phallicata narrow at base and slightly expanded at mid length, curved caudad for about 90° subapically beyond pair of short subapicodorsal lobes and apical hook directed dorsad.

Female. Unknown.

Etymology. An adjective in nominative singular from “She-xian,” a county in Anhui Province, China, referring to the type locality of this species.

Distribution. This species has been found only at the type localities in She County, An-hui Province, east central China, Oriental Region.

***Metalype truncata* Qiu & Morse, sp. n.**

<http://zoobank.org/E51038FF-C1F6-4F79-9E2B-3F999021F30B>

Fig. 4A–E

Metalype truncata Li, 1998: 221, figs 11.9–11.12, *nomen nudum*.

Type locality. PRC, Si-chuan Province: Jiu-zhai-gou National Park, Jiu-zhai-gou County, 33°16.02'N; 103°55.08'E, 2000 m, 25 June 1990, Collector Chen Xiao-en (Chen X-e); Du-jiang-yan City, Qing-cheng mountain, Wei-jiang River, 32 km SW of Du-jiang-yan downtown, 30°53.90'N; 103°34.37'E, 930 m, 20 June 1990, Collector JC Morse, Yang L-f, Li Y-w and Chen X-e,

Type specimen. Holotype. Male, in 75% ethanol, in cotton-stoppered microvial inside screwcap vial. Original label: “Sichuān, Jiǔ-zhài-gōu, Nán-píng-xiàn, 2000 M elev., 25 June, 1990, Coll. Chen” “*Metalype truncata*, Holotype, Li & Morse 1996”. Deposited in NJAU.

Paratypes. 19 males, in 100% ethanol, one specimen in cotton-stoppered microvial with genitalia removed and cleared. Original label: “Sichuān, Qīng-chéng-shān, 32 KM S.W. of Guàn xiàn, Wèi-jiāng-hé, 900 M elev., 27 June, 1990, Coll. Morse, Yang, Li, Chen” “*Metalype truncata* sp. n., Paratype, Li & Morse 1996” “Si-chuan Province P.R.C. Wei-jiang River Qin-cheng-shan, 32 km SW. of Du-jiang-yan City” [Handwritten]. Deposited in CUAC.

4 males, “Si-chuan Province P.R.C. Wei-jiang River Qin-cheng-shan, 32 km SW. of Du-jiang-yan City. Coll. Chen” [Handwritten]. Deposited in NJAU.

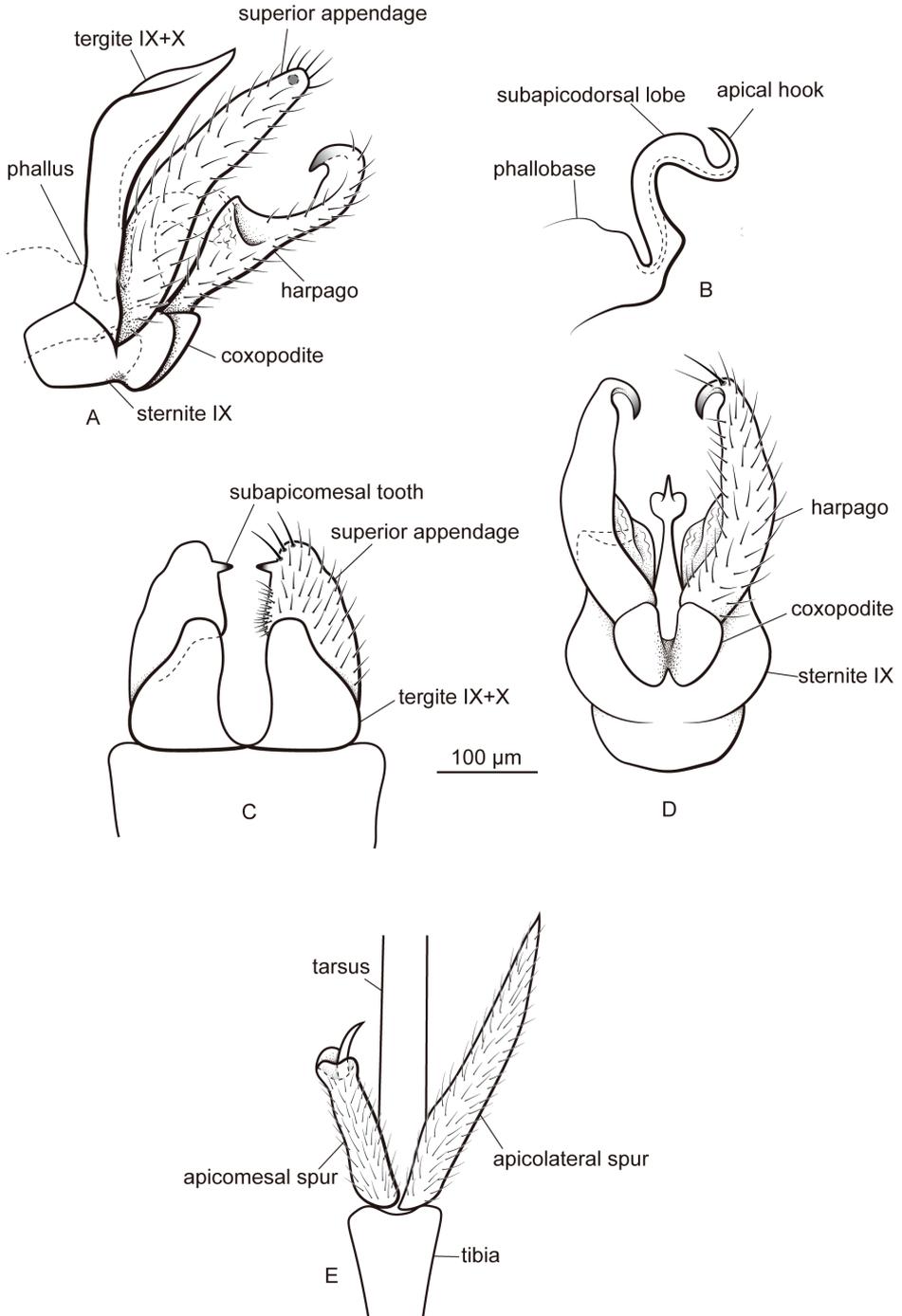


Figure 4. Male of *Metalype truncata* sp. n. **A** genitalia, left lateral **B** phallus, left lateral **C** genitalia, dorsal **D** genitalia, ventral **E** apical spurs of right hind leg, ventral.

Diagnosis. This species resembles *Metatype hubeiensis* sp. n. The differences are as detailed above for the latter species.

Description. Male. Forewings each 3.9–4.5 mm (n = 10). Compound eyes black, body light brown. Apicomeral spur of each hind tibia truncate apically, with lobes on edge and central acute process.

Genitalia. In lateral view tergites IX+X slightly concave dorsally and acute at apex, in dorsal view each half round at apex. In dorsal view superior appendages setose, each with mesal setae short and apical setae thicker; in lateral view digitate, slightly curved caudad at mid length and gradually narrowed to blunt apex, in dorsal view subtriangular, each with subapicomeral tooth about 1.5 times as long as wide. In ventral view sternite IX slightly expanded posteriorly. In ventral view coxopodites ovate, fused for about half of their length, in lateral view triangular. In lateral view harpagones narrow at bases, gradually expanded to mid length, then narrowed abruptly, with dorsal surface of expanding area weakly sclerotized and slightly concave posteriorly; in ventral view harpagones hooked mesodorsad at apex, apex sclerotized and recurved anterad. Phallobase expanded, phallicata with small basoventral corner, then strongly sinuous and curved at mid length about 100°, with pair of wide subapicodorsal lobes, hooked about 170° dorsad apically.

Female. Unknown.

Etymology. A Latin adjective in nominative singular, *truncata*, English “truncate,” referring to the apicomeral spur on each hind tibia.

Distribution. This species has been found only in the type localities in Si-chuan Province, central China, Oriental Region.

***Metatype uncatissima* (Botosaneanu, 1970), new record**

Fig. 5A–E

Psychomyia uncatissima Botosaneanu, 1970: 301–302. Type Locality: North Korea (Hamgiǒng-pukto); Levanidova et al. 1995: 7; Mey and Nozaki 2006: 24.

Metatype uncatissima (Botosaneanu, 1970): Li and Morse 1997: 274–275; Nozaki and Nakamura 2007: 94; Ivanov 2011: 191; Torii 2011: 7–12; Torii and Nakamura 2016: 425, 427, 429.

Material examined. 54 males, in 80% ethanol. One in cotton-stoppered microvial inside screwcap vial, with genitalia removed and cleared. Original label: “Heilongjiang, Shangzhixian, Maoershan-Town, Ashi River, Elev. 300 M, July 13, 1993, coll. Li Youwen & Sun Changhai” “*Metatype uncatissima*, (Botosaneanu)”. (50 in CUAC, 4 in NJAU).

Distribution. This species has been reported from North Korea, Japan, and the Russian Far East. We report it now also from northeastern China (Hei-long-jiang Province), East Palearctic Region. The collection sites are: PRC, Hei-long-jiang Province: Shang-zhi County, Mao-er-shan Town, A-shi River, 45°16.40'N; 127°30.26'E,

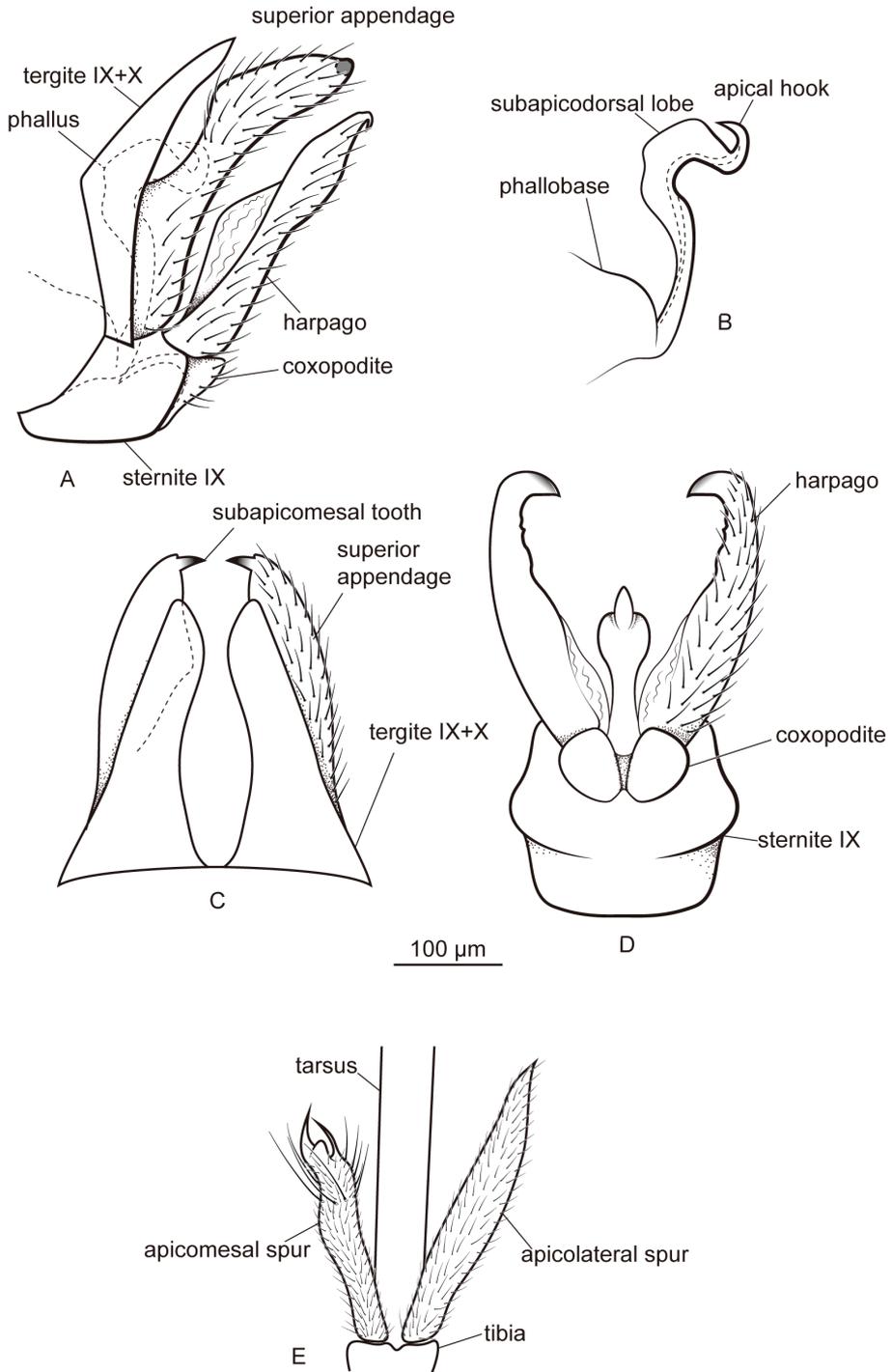


Figure 5. Male of *Metatypa uncatissima* (Botosaneanu, 1970). **A** genitalia, left lateral **B** phallus, left lateral **C** genitalia, dorsal **D** genitalia, ventral **E** apical spurs of right hind leg, ventral.

300 m, 13 July 1993, Collector Li Y-w and Sun C-h, 54 males (50 in CUAC, 4 in NJAU); Shang-zhi County, Wei-he Town, Yu-ling Tree Farm, close to Niu-shan Bridge, 44°39.33'N; 128°13.90'E, 380 m, 13 July 1993, coll. Li Y-w and Sun C-h, 1 male (NJAU); Tie-li City, Lang-xiang Town, Bei-lan River, Ba-lan Farm, 46°37.58'N; 129°7.29'E, 160m, 5 August 1993, coll. Li Y-w and Sun C-h, 2 males (NJAU); Wu-yi-lin Town, Yong-sheng, Wu-yun River, 48°37.09'N; 129°32.96'E, 160 m, 31 Jul. 1993, coll. Sun C-h, 4 males (NJAU).

In addition to the characters mentioned in the original description for this species (Botosaneanu 1970), the male apicomeresal spur of each hind tibia is slightly twisted, bearing a transverse row of setae subapically (Fig. 5E); the apex has two acute processes and a short hump. The female was illustrated by Li and Morse (1997).

Discussion

To date, only the characters of the type species, *Metalype fragilis*, have been used to diagnose the genus *Metalype*. Among the diagnostic characters now known to distinguish *Metalype* and *Paduniella*, synapomorphic characters for *Metalype* include the apicomeresal spurs of the hind tibiae that are short and curved, twisted, truncate or forked apically; in the male genitalia the subapicomeresal teeth of the superior appendages and the contorted phallus without a paramere. Synapomorphic characters for *Paduniella* include the 6-segmented maxillary palps, 4-segmented labial palps, and compressed male harpagones (Li and Morse 1997).

According to Li and Morse (1997, 1998), the most obvious differences between males of *Metalype* and *Psychomyia* are (1) The presence or absence of subapicomeresal teeth on the superior appendages; (2) the size of the mesodorsal expansion of the basal half of each harpago; (3) the presence or absence of membranous basodorsal surfaces of the harpagones; and (4) the degree of fusion of male tergites XI+X with the superior appendages. These and other characters and their polarities are indicated in Table 2.

The presence or absence of subapicomeresal teeth on the superior appendages is easily recognized. However, similar teeth are found in *Psychomyia amor* Malicky and Chantaramongkol, 1997; *P. amphiaras* Malicky and Chantaramongkol, 1997; *P. andromache* Malicky, 1997; *P. andromeda* Malicky, 1997; *P. asvagosha* Schmid, 1961; *P. capillata* Ulmer, 1910; *P. dasaratha* Malicky, 1993b; *P. holzenthali* Schmid, 1997; *P. kalais* Malicky, 2004b; *P. kiskinda* Malicky and Chantaramongkol, 1993; *P. klapaleki*, *P. kumari* Schmid, 1997; *P. kuni* Malicky and Chantaramongkol, 1993; *P. lak* Malicky and Chantaramongkol, 1993; *P. monto* Malicky and Chantaramongkol, 1993; *P. nevoissi* Schmid, 1997; *P. nithaiab*, *P. sinon* Malicky and Prommi, 2006; *P. sonlana* Oláh and Malicky, 2010; *P. vietnama* Oláh and Malicky, 2010; and *P. wigginsii* Schmid, 1997; Among them, *P. nithaiab*, *P. holzenthali*, *P. kumari*, and *P. klapaleki* are very similar to the three species transferred to *Metalype* by Li and Morse (1997): *M. anaktujub*, *M. mahayinna* (Schmid, 1961), and *M. uncatissima*. The latter three species were included in Schmid's *Psychomyia mahayinna* group together with *Metalype*

Table 2. Characters of selected *Psychomyia* species and all *Metatype* species, including species transferred here to *Metatype* (*). **Bold** character states are apomorphic. W = width, L = length.

Species	Male superior appendages subapicomeral teeth	Male harpagones expanded dorsally	Male harpagones membranous dorsally	Male hind tibiae apicomeral spurs length	Male hind tibiae apicomeral spurs shape	Male tergites IX+X fused with superior appendages	Female transverse row of setae on segment IX	Larval submental sclerites	Larval ventral apotome
<i>M. fragilis</i>	with	yes	yes	mesal>lateral	curved	no	with	W>L	W>5L
<i>M. anakrujuh</i>	with	yes	?	?	?	no	?	?	?
<i>M. bubeiensis</i>	with	yes	yes	mesal>lateral	forked, curved	no	?	?	?
<i>M. mahayinna</i>	with	yes	?	mesal>lateral	truncate	no	?	?	?
<i>M. shexianensis</i>	with	yes	yes	mesal>lateral	forked, curved	no	?	?	?
<i>M. truncata</i>	with	yes	yes	mesal>lateral	truncate	no	?	?	?
<i>M. uncatissima</i>	with	yes	yes	mesal>lateral	forked, twisted	no	with	W>L	W>5L
<i>M. holzenbali*</i>	with	yes	?	?	?	no	?	?	?
<i>M. kalpalakti*</i>	with	yes	yes	mesal>lateral	curved	no	?	W>L	W>5L
<i>M. kumari*</i>	with	yes	?	?	?	no	?	?	?
<i>M. nithaiab*</i>	with	yes	yes	mesal>lateral	truncate	no	?	?	?
<i>P. flavida</i>	without	no	no	lateral>mesal	straight, acute	yes	without	L>W	W<2L
<i>P. pusilla</i>	without	no	no	lateral>mesal	straight, acute	yes	without	L>W	W<2L
<i>P. nomada</i>	without	no	no	lateral>mesal	straight acute	yes	?	L>W	W<2L

fragilis (Schmid 1997), so we hypothesize that these four species also belong to the genus *Metalype* and we cite them in *Metalype* through the remainder of this paper. All of the other 17 species above with subapicomesal teeth on the superior appendages are very different from *Metalype* by the following characters: (1) Tergites IX+X are fused with the superior appendages completely (synapomorphy; tergites IX+X are separated from the superior appendages in *Metalype*); (2) the superior appendages are greatly expanded basodorsally (synapomorphy; the superior appendages are not expanded in *Metalype*); (3) the superior appendages each have a large mesal concavity (synapomorphy; the superior appendages are without concavities in *Metalype*); (4) the coxopodites are semicircular (semicircular condition is synapomorphic; the coxopodites are round, triangular, or rectangular in *Metalype*); (5) the harpagones are forked (synapomorphy; the harpagones are single in *Metalype*).

The phallicata is more or less vertical basally and has a reversed-S-shape with an apical hook directed dorsad in *Metalype* species and all of these 17 *Psychomyia* species. This general shape is a synapomorphy for *Psychomyia* and *Metalype*, with the phallicata of *Psychomyia* species other than those 17 species generally more nearly horizontal and evenly curved, probably apomorphic within *Psychomyia*.

Moreover, *P. sonlana*, *P. sinon* and *P. andromache* also have a few more mesal spines on the superior appendages. Considering that there are many *Psychomyia* species with dense spines on the mesal surfaces of the superior appendages, it is possible that the subapicomesal teeth in these species are remnants or a modification of the mesal spines in one or more monophyletic groups within genus *Psychomyia* and thus these spines are a homoplasy, not homologous with the synapomorphic subapicomesal teeth of *Metalype*.

The peculiar shape of the expansion of the harpagones is not observed in *Psychomyia* species. It is not apparent also in *Metalype shexianensis* and *M. anaktujuh*. Instead, these two species have a mesal process on each of harpago, possibly representing a dorsal hump that shifted apicomesad. *Metalype holzenthali*, *M. klapaleki*, and *M. nithaiah* have that kind of expansion; whereas *M. kumari* has mesal processes that resemble those of *M. shexianensis* and *M. anaktujuh*. This expansion, possibly modified into a mesal process in some species, is likely a synapomorphy for some, if not all species of *Metalype*.

The membranous basodorsal surfaces of the harpagones are present in all *Metalype* specimens we observed, but this character is seldom mentioned in descriptions. Botosaneanu (1970) described this character in the original description of *Metalype uncatisima*. Malicky (1995a) mentioned this character in his re-description of *M. fragilis* and his description of *M. klapaleki*. Under the dissecting microscope, the membranous part is often without setae, the color is white or light yellow, the boundary between the membranous and the non-membranous parts is very obvious; after clearing, the membranous part is transparent and almost invisible, so that it can be distinguished from other parts. This character is likely a synapomorphy for *Metalype*.

Schmid (1997) described the separation of tergites IX+X and superior appendages as a character of his *Psychomyia mahayinna* group. This separation can be recognized in all *Metalype* species and the fusion of these structures is seen in most *Psychomyia* species. However, the fusion of tergites IX+X with the superior appendages is not very

obvious in some *Psychomyia* species, for example *Psychomyia arefinae* Schmid, 1997; *P. schefferae* Schmid, 1997 and *P. scottae* Schmid, 1997. On the other hand, the bases of the superior appendages can be very wide (*Metalype anaktujuh*, *M. shexianensis*), which can make this character ambiguous. *Metalype holzenthali*, *M. klapaleki*, *M. kumari*, and *M. nithaiah* all have tergites IX+X separated from the superior appendages, as for other *Metalype* species. Thus the fusion of tergites IX+X and superior appendages seems to be a synapomorphy within genus *Psychomyia*.

The apicomesal spurs of hind tibiae on Psychomyiidae species other than those of *Metalype* are straight and acute. On all *Metalype* species we have studied, the apicomesal spurs are shorter than the apicolateral spurs, and these apicomesal spurs are more or less curved, twisted, truncate, or forked apically (Figs 2E, 3F, 4E, 5E, 6). All *Psychomyia* specimens we observed (including *Psychomyia flavida* Hagen, 1861; *P. extensa* Li, Sun, and Yang, 1999; *P. nomada* (Ross, 1938), and eleven unpublished species from China) have apicomesal spurs straight and slightly longer than the apicolateral spurs, never forked or truncate. *Metalype mahayinna* has apicomesal spurs similar to those of *Metalype truncata* (Malicky 1996; pers. comm). Males of *M. nithaiah* and *M. klapaleki* (Fig. 7) also have the apicomesal spur on each hind tibia shorter than the apicolateral spur and curved apically, supporting the hypothesis that these species belong in *Metalype*. The spurs of *M. holzenthali* and *M. kumari* are unknown to us. The slightly curved, twisted, forked or truncate apicomesal spurs on male hind tibiae is a synapomorphy within genus *Metalype*.

A difference between *Metalype* and *Psychomyia* females is that those of *Metalype* have a transverse row of setae on segment IX and those of *Psychomyia* species are without these setae (presence of the transverse setal row is synapomorphic). This difference is observed in females of *Psychomyia usuguronis* (Matsumura, 1931) (Ito et al. 2011), *P. flavida* (Ito et al. 2000), *P. pusilla* (Malicky 2004a), *M. fragilis* (Malicky 2004a), and *M. uncatissima* (Li and Morse 1997). However, the females are unknown for the four *Psychomyia* species we hypothesize here to belong to *Metalype* (*M. nithaiah*, *M. holzenthali*, *M. kumari*, and *M. klapaleki*).

Edington and Hildrew (1995) compared the larvae of *Psychomyia pusilla* and *Metalype fragilis*, and found three differences between them: (1) *Psychomyia pusilla* has the submental sclerites longer than wide, with dark patterns (synapomorphy); *M. fragilis* has the sclerites wider than long and without patterns. (2) *Psychomyia pusilla* has the ventral apotome small and triangular, no more than two times as wide as long (synapomorphy); *M. fragilis* has the ventral apotome expanded laterally, more than five times as wide as long. (3) *Psychomyia pusilla* has five or six teeth on the mesal surface of each anal claw; *M. fragilis* has two or three teeth (character polarity uncertain).

The long submental sclerite character is found in *Psychomyia flavida* and has been used for distinguishing the larvae of *Psychomyia* and *Paduniella*, with these sclerites wider than long in the latter (Wiggins 1996, Li and Morse 1997, 1998, Morse and Holzenthal 2008). We observed this long submental sclerite character for *P. nomada* specimens in the CUAC. On the other hand, the wide submental sclerites on larvae of *Metalype* species have been confirmed for the larvae of *M. fragilis* and *M. uncatissima*

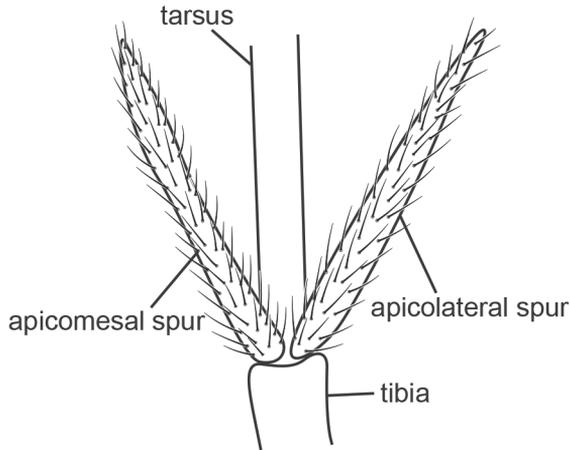


Figure 6. Male of *Metalype fragilis* (Pictet, 1834). Apical spurs of right hind leg, ventral.

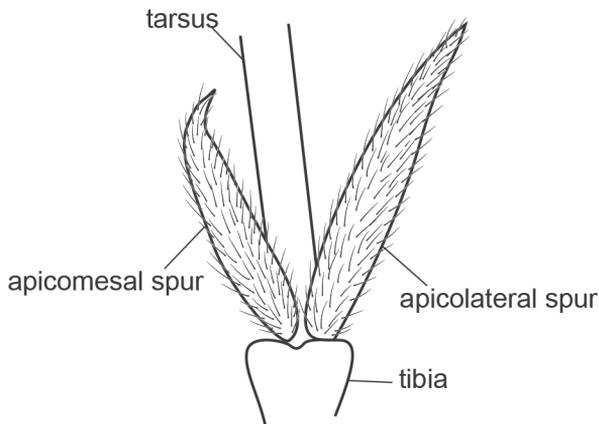


Figure 7. Male of *Metalype klapaleki* (Malicky, 1995a). Apical spurs of right hind leg, ventral.

by many authors (Waringer and Graf 2011, Urbanič et al. 2003, Coppa et al. 2009, Torii 2011, Torii and Nakamura 2016). For all the species mentioned above, the small ventral apotome is usually coupled with the longer submental sclerites. One exception is the *Psychomyia* sp. larva from Aichi (Torii and Nakamura 2016); that larva has submental sclerites longer than their width, but the ventral apotome is wide. Dark patterns on the submental sclerites of *Psychomyia* are always present, although sometimes faint.

Coppa et al. (2009) concluded that the main character distinguishing the larva of *Paduniella vandeli* Decamps, 1965 from that of *Metalype fragilis* is the number of teeth on the ventral margin of each anal claw. The final instar larva of *P. vandeli* bears seven or eight teeth on each anal claw (Coppa et al. 2009) while that of *M. fragilis* bears only two or three teeth (Coppa et al. 2009, Edington and Hildrew 1995). On

the other hand, the larva of *M. uncatissima* has eight teeth on each anal claw (Torii 2011), *Paduniella nearctica* Flint, 1967 has four to six teeth; *Psychomyia flavida* (Morse and Holzenthal 2008) and *Psychomyia* sp. (probably *P. lumina*, Wiggins 1996) each have four teeth, and *P. nomada* has three or four teeth. The third instar larva of *P. vandeli* also has three teeth on each anal claw (Coppa et al. 2009). Moreover, the teeth may not be uniform; some of them can be very small and hard to recognize. Thus, the number of teeth on each anal claw is not a reliable character for distinguishing the three genera.

Torii and Nakamura (2016) identified larvae of Psychomyiidae by molecular methods. They compared the morphological characters of larvae and noted that the episternum of each foreleg of *Metalype uncatissima* is without a vertical suture while larvae of *Paduniella horaiensis* Nishimoto, 2011 and *Psychomyia* sp. have the suture. We observed this suture on *P. nomada* specimens, but it is also present on the larva of *M. fragilis* (Coppa et al. 2009), so that the absence of the suture may be an autapomorphy of *M. uncatissima*. Torii and Nakamura (2016) also mentioned that the mature larva of *Metalype* (5–6 mm) is longer than the larva of *Paduniella* (3–4 mm). The phylogenetic evidence and diagnostic differences for larvae of *Metalype* and *Paduniella* remain inconclusive until more information on larvae is available.

The larva of *M. klapaleki* has submental sclerites wider than long. In fact, no differences have been found between larvae of *M. klapaleki* and larvae of *M. fragilis* (Urbanič et al. 2003), further supporting our hypothesis that *M. klapaleki* is a species of *Metalype*. Larvae are unknown for the other three species that we transfer here to *Metalype*. When they become known, we predict that the larval characters for those species will support our hypothesis.

Conclusion

The male genitalia of *Metalype* and *Psychomyia* are very similar to each other, but there are some distinctive characters supporting the monophyly of each genus. The details are shown in Table 2. The known female genitalia and larvae of *Metalype* are similar to those of *Paduniella* and both of them are very different from female genitalia and larvae of *Psychomyia*. Treating *Metalype* as a synonym of *Psychomyia* may cause difficulties for identifying females and larvae of *Psychomyia*. However, female genitalia and larvae of only a few species are known in these genera, such that more information will be helpful. Based on the characters of males, we conclude that the following species should be transferred from *Psychomyia* to *Metalype*:

Metalype holzenthali (Schmid, 1997), comb. n.

Metalype klapaleki (Malicky, 1995a), comb. n.

Metalype kumari (Schmid, 1997), comb. n.

Metalype nithaiah (Malicky, 2014), comb. n.

Key to males of *Metatype* species

- 1 Superior appendages each with subapicomesal tooth and with tergites IX+X separated from superior appendages; hind tibiae each with apicomesal spur shorter than apicolateral spur and more or less curved, twisted, truncate, or forked apically ***Metatype***
- Superior appendages usually without subapicomesal teeth and with tergites IX+X fused with superior appendages; hind tibiae each with apicomesal spur longer than apicolateral spur, straight, and acute apically ***Psychomyia***
- 2 Harpagones in ventral view each with large subapicomesal process, as large as apex (Fig. 3C)..... **3**
- Harpagones in ventral view without large subapicomesal processes (Figs 2D, 4D, 5D)..... **5**
- 3 Harpagones in ventral view each with small mesal process behind larger mesal process (Fig. 3C)..... ***M. shexianensis***
- Harpagones in ventral view without small mesal processes **4**
- 4 Halves of tergites IX+X in dorsal view separated widely from each other, more than twice width of each base (Schmid 1997, fig. 17) ***M. kumari***
- Halves of tergites IX+X in dorsal view separated narrowly from each other, separation about as much as width of each base (Malicky 1995b, page 23, figs on the top right corner) ***M. anaktujub***
- 5 Harpagones in ventral view not hooked mesad apically (Schmid 1961, pl. 15, fig. 1)..... ***M. mahayinna***
- Harpagones in ventral view each hooked mesad apically (Figs 2D, 4D, 5D) ... **6**
- 6 Halves of tergites IX+X in dorsal view round apically (Fig. 4C) **7**
- Halves of tergites IX+X in dorsal view attenuate and blunt apically (Figs 2C, 5C) **9**
- 7 Harpagones in ventral view each strongly hooked, with apices recurved anterad (Fig. 4D)..... ***M. truncata***
- Harpagones in ventral view not as strongly hooked, with apices pointing mesad (Fig. 5D)..... **8**
- 8 Harpagones in lateral view longer than superior appendages (Schmid 1997, fig. 12), Oriental Region..... ***M. holzenthali***
- Harpagones in lateral view shorter than superior appendages (Malicky 1995a, fig. 1), West Palearctic Region..... ***M. kalpaleki***
- 9 Halves of tergites IX+X in dorsal view curved slightly laterad apically (Malicky, 2014, pl. 8)..... ***M. nithaiab***
- Halves of tergites IX+X in dorsal view not curved laterad apically (Figs 2C, 5C) **10**
- 10 Superior appendages in dorsal view each strongly narrowed in apical half (Fig. 2C) ***M. hubeiensis***
- Superior appendages in dorsal view not narrowed at apical half (Fig. 5C) ... **11**

- 11 Harpagones in ventral view each expanded near bases (Fig. 5D), East Palearctic Region *M. uncatissima*
 – Harpagones in ventral view expanded near mid length (Malicky 1995a, fig. 1), West Palearctic Region *M. fragilis*

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DNA barcoding and species delimitation of Chaitophorinae (Hemiptera, Aphididae)

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Abstract

Chaitophorinae aphids are widespread across Eurasia and North America, and include some important agricultural and horticultural pests. So, accurate rapid species identification is very important. Here, we used three mitochondrial genes and one endosymbiont gene to calculate and analyze the genetic distances within different datasets. For species delimitation, two distance-based methods were employed, threshold with NJ (neighbor-joining) and ABGD (Automatic Barcode Gap Discovery), and two tree-based approaches, GMYC (General Mixed Yule Coalescent) and PTP (Poisson Tree Process). The genetic interspecific divergence was clearly larger than the intraspecific divergence for four molecular markers. COI and COII genes were found to be more suitable for Chaitophorinae DNA barcoding. For species delimitation, at least one distance-based method combined with one tree-based method would be preferable. Based on the data for *Chaitophorus saliniger* and *Laingia psammae*, DNA barcoding may also reveal geographical variation.

Keywords

Chaitophorinae, distance-based analysis, gnd, mitochondrial genes, tree-based analysis

Introduction

Aphids from more than 5,000 species (Favret 2016) feed on plant phloem directly and spread various plant diseases (Blackman and Eastop 2000), many serving as important economic pests. The identification of aphid species based on morphological characteristics faces tremendous challenges due to their complicated life cycle, polymorphism, phenotypic plasticity, and numerous morphs (Zhang and Zhong 1983, Foottit et al. 2009a). Chaitophorinae lies within Aphididae, and comprises two tribes, Chaitophorini and Siphini, including 196 species and subspecies in 12 genera (Remaudiere and Remaudiere 1997, Favret 2016). The subfamily is distributed mainly in the Palaearctic (about 80% of species), and Nearctic (Richards 1972, Qiao 1996, Liu et al. 2009, Wieczorek 2010). Most species in this subfamily are monoecious holocyclic, but some species, such as *Sipha* (*Sipha*) *flava* and *Sipha* (*Rungsia*) *maydis*, may be anholocyclic in regions with milder winters (Blackman and Eastop 2000, Wieczorek 2010, Wieczorek and Bugaj-Nawrocka 2014). The Chaitophorini is mainly associated with plants of the families Salicaceae and Aceraceae (Blackman and Eastop 1994), whereas the Siphini infest plants in the Poaceae, Cyperaceae, Juncaceae and Typhaceae (Blackman and Eastop 2006). Additionally, individual species often have high host specificity (Blackman and Eastop 1994). Species identification of Chaitophorinae aphids can be difficult when based on their morphological characteristics. The Chaitophorini have clear morphological differences between genera, but *Chaitophorus* (109 known species) and *Periphyllus* (49 known species) have high species diversity (Essig 1952, Hille Ris Lambers 1960, Pintera 1987, Qiao 1996); and the morphological differences between species within these genera are relatively slight, often depending on the chaetotaxy of the body dorsum and appendages (Pintera 1987). In the Siphini, both among genera and between species, overlap and convergence of morphological characteristics are common, and genus and species identification are not easy. *Sipha* in particular (11 known species) has relatively great diversity, and species identification can be a problem.

DNA barcoding based on a short fragment of mitochondrial DNA can provide an effective tool for species diagnosis. In animals, the 5' end of mitochondrial cytochrome c oxidase I (COI) with a 658-bp fragment was selected as a standard DNA barcode (Hebert et al. 2003). This has been widely used for identifying unknown specimens and the rapid identification of species (Hebert et al. 2003, Wang and Qiao 2009, Wang et al. 2013b, Wen et al. 2013). Its practicability and effectiveness have been recognized and accepted in some insect groups, such as Diptera (Scheffer et al. 2006), Lepidoptera (Hajibabaei et al. 2006), Ephemeroptera (Ball et al. 2005), Hemiptera (Lee et al. 2011), Coleoptera (Lobl and Leschen 2005), and Hymenoptera (Smith et al. 2008). Additionally, the application range was expanded to pest control and quarantine (Armstrong and Ball 2005, Ratnasingham and Hebert 2007, Naaum et al. 2012, Pelletier et al. 2012). For aphids, the DNA barcoding approach has played an efficient role in the rapid identification of species on specific plants (Naaum et al. 2012, Chen et al. 2013b, Wang et al. 2013a, Wang et al. 2013b, Wen et al. 2013,

Wang et al. 2015), the effective distinction of morphologically indistinguishable species and subspecies (Wang and Qiao 2009, Rebijith et al. 2013, Cocuzza and Cavalieri 2014, Beji et al. 2015, Kinyanjui et al. 2016), the recognition of cryptic species (Rebijith et al. 2013, Lee et al. 2015), and in species classification (Cocuzza and Cavalieri 2014, Mroz et al. 2015). Likewise, DNA barcoding may be used in species diversity assessment within different regions (Podmore et al. 2015, Chen et al. 2016); and is a powerful tool for the identification of multi-life stages, different morphs, and biological debris (Shufran and Puterka 2011). Crucially, it has improved the monitoring and control of pest aphids. The identification of aphid species is often difficult due to the shortage of easily distinguishable morphological characteristics, or feature convergence (Wang and Qiao 2009). Some chaitophorine species are important agricultural, forestry, and horticulture pests, for which accurate identification is necessary. At the authors' last count (2016.04.06), researchers have provided some chaitophorine DNA barcoding sequences for 36 species to the NCBI and for 49 species to the Barcode of Life Data System (BOLD) (Foottit et al. 2008, Foottit et al. 2009b, Lee et al. 2011, Gwiazdowski et al. 2015). However, the DNA barcoding of this group is insufficient. In this work, we sequenced 1,609 sequences from 670 samples in 8 genera from both tribes of Chaitophorinae, based on three genes from the aphid mitochondrial genome, and one from the endosymbiont *Buchnera*. We employed four methods (threshold with NJ, ABGD, GMYC, and PTP) to analyze sequence diversities and genetic divergences between different species and probe the efficiency of identifying species. Based on DNA barcoding data, we also discuss the influence of geographical distribution on population differentiation.

Materials and methods

Taxa sampling and gene selection

All samples were collected into and cryopreserved in 95% or 100% ethanol. DNA from one individual per sample was isolated for molecular studies and three to five individual aphids per collection were mounted on microscope slides for morphological examination. Preserved aphid colonies were examined prior to preparation to ensure that they did not consist of multiple species. Voucher specimens for each sample were identified by G.X. Qiao based on morphological diagnostic features using standard literature-based keys (esp. Blackman and Eastop 1994, Pintera 1987, Wieczorek 2010) and by a comparison with previously identified specimens in the National Zoological Museum of China, Beijing. To avoid mutual influence and to ensure the independence of the different research methods, the morphological identification and molecular research were performed independently. All samples and voucher specimens were deposited in the National Zoological Museum of China, Institute of Zoology, Chinese Academy of Sciences, Beijing, China. Details of the sequenced taxa and voucher information are listed in Suppl. material 1.

Three aphid genes were targeted: mitochondrial cytochrome oxidase c subunit I (COI), cytochrome oxidase c subunit II (COII), and cytochrome b (Cytb), and one aphid endosymbiont *Buchnera* gene gluconate-6-phosphate dehydrogenase (gnd) (Kim and Lee 2008, Wang and Qiao 2009, Zhang et al. 2011, Chen et al. 2012).

DNA extraction, amplification and sequencing

Total genomic DNA was extracted from single aphid. Individual aphids were selected from the ethanol-preserved candidates with a destructive DNA extraction procedure. Plump adults are the ideal experimental material, but they must be examined under a microscope (Leica DM 2500) to eliminate parasitized individuals. Total DNA was extracted by following the Quick-Start protocol of DNeasy Blood & Tissue Kit (QIAGEN, Dusseldorf, Germany) with a single individual. The DNA solution was then stored at -20 °C for subsequent molecular experiments.

The polymerase chain reaction (PCR) mixture for the amplification of COI, COII, Cytb, and gnd genes comprised 22 µl of double distilled water (ddH₂O), 3 µl of 10 × EasyTaq Buffer (+ Mg²⁺) (TransGen Biotech, Beijing, China), 2.4 µl of 2.5 mM/800 µl dNTPs (TransGen Biotech), 0.6 µl of 10 pmol/µl forward and reverse primers, 0.4 µl of 5 U/µl EasyTaq DNA Polymerase (TransGen Biotech), and 1 µl of DNA solution for a total volume of 30 µl.

The PCR conditions differed according to the gene and the specific primers, especially the annealing temperature, which was the most critical factor influencing product quality. The detailed primer information is shown in Suppl. material 2. The thermal setup of primer LepF/LepR (Footitt et al. 2008) or LCO1490/HCO2198 (Folmer et al. 1994) for COI gene fragment was: a 5-minute initial denaturation at 95 °C followed by 35 cycles of 30-second denaturation at 95 °C, 30 seconds of annealing at 50 °C, a 1-minute extension at 72 °C, and a 10-minute final extension at 72 °C. The protocol for primer mt2993+ (Stern 1994)/A3772 (Normark 1996) for tRNA/COII molecular marker was as follows: a 5-minute initial denaturation at 95 °C followed by 35 cycles of 1-minute denaturation at 95 °C, 1 min at 42 °C, 1 min at 72 °C, and a 7-minute final extension at 72 °C. The parameters of primer CP1/CP2 (Harry et al. 1998) for Cytb amplification was simplified as: 94 °C with 5 min, and 40 cycles of 94 °C with 50s, 48 °C with 1 min, 72 °C with 1.5 min, and 72 °C with 10 min. The setup of primer BamHI/ApaI (Clark et al. 1999) for *Buchnera* gnd gene was predigested as: 94 °C with 5 min, and 30 cycles of 94 °C with 1 min, 55 °C with 30s, 72 °C with 1 min, and 72 °C of 10 min final extension.

The amplification products were detected by 1.5% agarose gel electrophoresis (AGE), and then purified using EasyPure Quick Gel Extraction Kit (TransGen Biotech). The eligible products were then sent to TsingKe Biological Technology, Beijing, China or BGI, Shenzhen, China for sequencing, which was required to be bidirectional.

Sequence edition and alignment

The returned forward and reverse chromatograms were loaded and then assembled and edited by SeqMan in DNASTar software (DNASTAR, Madison, Wisconsin, USA). The nucleotide sequences were first examined in NCBI by Basic Local Alignment Search Tool (BLAST) (Altschul et al. 1990) to test their affiliations. Concurrently, for the encoding gene fragments, we translated the assembled contigs into amino acids by MEGA6 (Tamura et al. 2013) to examine whether the sequences were correct and accurate. Multiple alignments were accomplished by MAFFT (Katoh and Standley 2013), and the sequences were then adjusted and trimmed manually in MEGA6. It is noteworthy that the sequences amplified with primer mt2993+/A3772 covered the COII gene fragment as well as a tRNA, which was then removed for subsequent analysis.

Species delimitation methods

In addition to sequences from 425 samples, we downloaded 245 COI and 1 COII sequence from NCBI. Here, we defined the datasets as COI-670 (including the whole research group and NCBI sequences), COII-376 (including 375 internal sequences and 1 NCBI sequence), Cytb-413 (newly gotten for this study), gnd-396 (newly obtained sequences), and COI-338, COII-338, Cytb-338, gnd-338, which contained only the specimens that acquired all 4 gene sequences.

A neighbor-joining (NJ) (Saitou and Nei 1987) tree was constructed by MEGA6 based on the aligned sequences. One thousand bootstrap replications were calculated to assess the credibility of the NJ analysis. The Kimura 2-parameter (K2P) model of base substitution (Kimura 1980) was selected in pairwise distances calculation, and for the more accurate comparison between sequences, the pairwise deletion pattern was selected for gaps/missing data treatment. After bootstrap consensus trees with bootstrap values at each node were obtained, we computed the condensed tree with a 50% cut-off value for the consensus tree. When analyzing the COI-670 tree, we chose a threshold of 2% (Footitt et al. 2009b) for a cluster standard, which has been well used in aphids. With regard to the COII-376, COII-338, Cytb-413, Cytb-338, gnd-396 and gnd-338 NJ trees, we calculated only the cluster topologies.

The Automatic Barcode Gap Discovery (ABGD) (Puillandre et al. 2012a) approach is a model-based method for delimiting species. Based on the existence of a barcoding gap (namely the intraspecific divergences are smaller than interspecific divergences) and a prior intraspecific divergence (p), the ABGD procedure first sorts the dataset into a hypothetical species, and then computes recursively with the previous groups to obtain a result optimized until there are no better partitions. We ran the ABGD with a graphic web version (<http://wwwabi.snv.jussieu.fr/public/abgd/abgd-web.html>). First, we calculated the distance values among samples by using MEGA6 with p-distance, Jukes-Cantor (JC69) model, and K2P model separately, and the

result data were saved as CSV format file. We then chose 0.055, which had been suggested for Aphididae (Footitt et al. 2009b), as the prior intraspecific divergence for COI-670 and COI-338 datasets, and we used a p value of 0.1, which was the default and shown to be sufficient for analysis, for the other data sets. The other parameters were maintained by default for all analyses.

The General Mixed Yule Coalescent (GMYC) (Fujisawa and Barraclough 2013) is a tree-based approach for the delimitation of species. We ran the GMYC method in R project (available from: <https://www.r-project.org/>) by using the “splits” package (available from: <http://r-forge.r-project.org/projects/splits>). The input tree was required to be strictly ultrametric and bifurcating, which meant there was no zero-length branch. Here, we used a maximum likelihood (ML) tree as the input. Therefore, the haplotypes were calculated and generated by DnaSP (Librado and Rozas 2009), and an ML tree was constructed by RAxML (Stamatakis 2006) with haplotype data. Due to the ultrametric and bifurcating requirement, the ML tree was constructed with r8s (Sanderson 2003). The outcome tree modified by r8s was read into the “splits” R package, and the delimiting result was obtained with relevant commands.

The Poisson Tree Process (PTP) (Zhang et al. 2013) model is another tree-based method for inferring putative species. The PTP approach is a close relative of the GMYC method, but it only needs a simple phylogenetic tree as its input without requiring it to be ultrametric and bifurcating. As an updated version of the original PTP, the bPTP method was employed simultaneously to separate hypothetical species, which added a Bayesian support value to the tree. The PTP and bPTP analyses were run on a web server (<http://species.h-its.org/ptp/>) and the value 500,000 was selected for MCMC generations, with the other parameters set by default. The input tree was an ML tree constructed by RAxML with GTRCAT model. However, we encountered the same problem as Schwarzfeld and Sperling (2015), namely that the bPTP analysis failed to show convergence under 500,000 generations (the upper limit of the web server). Therefore, only the PTP result is displayed and discussed below.

Results

Morphological identification

The 425 samples collected by the group members in recent years were carefully authenticated with mounted individuals under the microscope, and all 425 samples were identified to species. The few vouchers with uncertain species identification were sorted into featured clusters and were given the epithet “sp.”, which made them convenient for further analysis. A total of 75 morphological species were determined from 670 whole samples, and 51 were identified from the 425 mounted samples.

Sequence alignment

The COI sequences were trimmed to a length of 658 bp, which included 365 conserved sites, 293 variable sites and 258 parsimony-informative sites. The sequences had an average nucleotide composition of 38.0% T, 17.1% C, 34.4% A, and 10.5% G. The COII sequences were trimmed to a final length of 672 bp, among which 399 sites were conserved, 273 sites were variable, and 251 sites were parsimony-informative. The average T, C, A, G compositions of these sequences were 38.7%, 14.0%, 39.5%, and 7.8%, respectively. The Cytb gene was 760 bp, in which there were 420 conserved sites, 340 variable sites and 303 parsimony-informative sites. The Cytb sequences consisted of 41.4% T, 15.3% C, 34.3% A, and 9.0% G. We obtained a total length of 807 bp for the *gnd* gene with an average nucleotide composition of 37.8% T, 9.8% C, 39.5% A, and 12.8% G, among which there were 368 conserved sites, 439 variable sites and 417 parsimony-informative sites. Across all 4 genes, a strong T and A nucleotide composition bias existed.

From a total of 425 samples, 425 COI gene fragment sequences, 375 COII gene fragment sequences, 413 Cytb gene fragment sequences, and 396 *gnd* gene fragment sequences were acquired. The successive amplification efficiency of those markers in order was COI (100%) > Cytb (97%) > *gnd* (93%) > COII (88%).

Genetic divergence analysis

Genetic divergences were assessed by 5 disparate metrics among and within species. For the interspecific divergences of congeneric species, we chose the average interspecific distance, which was calculated within genera that contained more than one species, and the smallest interspecific distance, which meant the minimal value of interspecific distance within genera with at least two species. When evaluating the intraspecific divergences, three variables (average intraspecific distance, mean theta, and average coalescent depth) were applied. The average intraspecific distance was the average value of the genetic distances between samples within species that had at least two individuals. The mean theta signified a modified theta, which expressed the average pairwise distance scored for species with more than one obtained representative, by dislodging improper individuals concerned with the asymmetrical acquisition of samples. The average coalescent depth, namely the average value of maximum intraspecific distance, was calculated for species in which there were no fewer than two samples.

All five interspecific and intraspecific metrics were determined within genera and species (Table 1). The results of different genes and datasets showed distinctly high interspecific divergences and low intraspecific distances. For DNA barcoding the smallest interspecific distance and average coalescent depth were the most useful and intuitive parameters. Within *Chaitophorinae*, the genetic divergence ranges of smallest interspecific distance and average coalescent depth of COI, COII, Cytb, and *gnd*

Table 1. The inter- and intra-specific genetic distances of congeneric species of Chaitophorinae.

Genus/Dataset (no. species/specimens)	Interspecific Distance		Intraspecific Distance		
	average interspecific distance	smallest interspecific distance	average intraspecific distance	mean theta	average coalescent depth
<i>Chaitophorus</i>					
COI-670(38/534)	0.1158±0.0191	0.1015±0.0178	0.0070±0.0060	0.0083±0.0057	0.0126±0.0126
COII-376(25/283)	0.0956±0.0246	0.0853±0.0207	0.0017±0.0019	0.0025±0.0019	0.0060±0.0054
Cytb-413(25/323)	0.1233±0.0281	0.0971±0.0260	0.0049±0.0066	0.0058±0.0068	0.0219±0.0357
gnd-396(25/306)	0.0996±0.0316	0.0807±0.0248	0.0020±0.0030	0.0034±0.0032	0.0042±0.0051
COI-338(25/253)	0.1117±0.0286	0.0995±0.0215	0.0058±0.0044	0.0077±0.0033	0.0088±0.0071
COII-338(25/253)	0.0950±0.0247	0.0855±0.0208	0.0018±0.0021	0.0027±0.0021	0.0062±0.0055
Cytb-338(25/253)	0.1169±0.0310	0.0983±0.0270	0.0043±0.0064	0.0052±0.0067	0.0164±0.0337
gnd-338(25/253)	0.0843±0.0264	0.0811±0.0255	0.0014±0.0016	0.0025±0.0014	0.0032±0.0033
<i>Lambersaphis</i>					
COI-670(1/3)	-	-	0.0040±0.0028	0.0060±0.0000	0.0060±0.0000
COII-376(1/3)	-	-	0.0047±0.0033	0.0070±0.0000	0.0070±0.0000
Cytb-413(1/3)	-	-	0.0053±0.0012	0.0053±0.0012	0.0070±0.0000
gnd-396(1/3)	-	-	0.0007±0.0005	0.0010±0.0000	0.0010±0.0000
COI-338(1/3)	-	-	0.0040±0.0028	0.0060±0.0000	0.0060±0.0000
COII-338(1/3)	-	-	0.0047±0.0033	0.0070±0.0000	0.0070±0.0000
Cytb-338(1/3)	-	-	0.0053±0.0012	0.0053±0.0012	0.0070±0.0000
gnd-338(1/3)	-	-	0.0007±0.0005	0.0010±0.0000	0.0010±0.0000
<i>Periphyllus</i>					
COI-670(19/83)	0.1113±0.0231	0.1075±0.0220	0.0040±0.0146	0.0080±0.0198	0.0218±0.0439
COII-376(13/53)	0.0936±0.0299	0.0938±0.0282	0.0007±0.0014	0.0027±0.0015	0.0024±0.0024
Cytb-413(14/54)	0.0975±0.0200	0.0944±0.0194	0.0020±0.0029	0.0041±0.0030	0.0052±0.0032
gnd-396(14/54)	0.1256±0.0669	0.1292±0.0602	0.0004±0.0010	0.0016±0.0014	0.0007±0.0012
COI-338(13/53)	0.0971±0.0258	0.0985±0.0248	0.0019±0.0032	0.0056±0.0032	0.0044±0.0035
COII-338(13/53)	0.0936±0.0299	0.0938±0.0281	0.0007±0.0014	0.0027±0.0015	0.0024±0.0024
Cytb-338(13/53)	0.0974±0.0203	0.0935±0.0206	0.0020±0.0029	0.0041±0.0030	0.0052±0.0032
gnd-338(13/53)	0.1250±0.0679	0.1283±0.0632	0.0004±0.0010	0.0016±0.0014	0.0007±0.0012
<i>Trichaitophorus</i>					
COI-670(3/3)	0.1233±0.0200	0.1233±0.0200	-	-	-
COII-376(3/3)	0.1103±0.0190	0.1103±0.0190	-	-	-
Cytb-413(2/2)	0.1040±0.0000	0.1040±0.0000	-	-	-
gnd-396(3/3)	0.1427±0.0162	0.1427±0.0162	-	-	-
COI-338(2/2)	0.0990±0.0000	0.0990±0.0000	-	-	-
COII-338(2/2)	0.1190±0.0000	0.1190±0.0000	-	-	-
Cytb-338(2/2)	0.1040±0.0000	0.1040±0.0000	-	-	-
gnd-338(2/2)	0.1600±0.0000	0.1600±0.0000	-	-	-
<i>Yamatochaitophorus</i>					
COI-670(3/3)	0.0043±0.0009	0.0043±0.0009	-	-	-
COII-376(3/3)	0.0037±0.0021	0.0037±0.0021	-	-	-
gnd-396(3/3)	0.0007±0.0005	0.0007±0.0005	-	-	-
<i>Chaetosiphella</i>					
COI-670(3/24)	0.0515±0.0418	0.0693±0.0490	0.0149±0.0128	0.0197±0.0111	0.0185±0.0165
COII-376(3/24)	0.0372±0.0368	0.0563±0.0399	0.0083±0.0051	0.0091±0.0047	0.0090±0.0090

Genus/Dataset (no. species/specimens)	Interspecific Distance		Intraspecific Distance		
	average interspecific distance	smallest interspecific distance	average intraspecific distance	mean theta	average coalescent depth
Cytb-413(3/24)	0.0481±0.0455	0.0703±0.0498	0.0140±0.0123	0.0154±0.0120	0.0230±0.0220
gnd-396(3/23)	0.0521±0.0608	0.0887±0.0627	0.0084±0.0068	0.0107±0.0058	0.0090±0.0090
COI-338(3/23)	0.0512±0.0422	0.0693±0.0490	0.0147±0.0128	0.0202±0.0107	0.0165±0.0145
COII-338(3/23)	0.0371±0.0369	0.0563±0.0399	0.0083±0.0052	0.0091±0.0048	0.0090±0.0090
Cytb-338(3/23)	0.0480±0.0457	0.0703±0.0498	0.0142±0.0124	0.0158±0.0121	0.0230±0.0220
gnd-338(3/23)	0.0521±0.0608	0.0887±0.0627	0.0084±0.0068	0.0107±0.0058	0.0090±0.0090
<i>Laingia</i>					
COI-670(1/2)	-	-	0.0640±0.0000	0.0640±0.0000	0.0640±0.0000
COII-376(1/2)	-	-	0.0680±0.0000	0.0680±0.0000	0.0680±0.0000
Cytb-413(1/2)	-	-	0.0620±0.0000	0.0620±0.0000	0.0620±0.0000
<i>Sipha</i>					
COI-670(5/17)	0.0940±0.0250	0.0882±0.0320	0.0082±0.0127	0.0147±0.0139	0.0118±0.0159
COII-376(2/5)	0.1115±0.0009	0.1110±0.0000	0.0027±0.0012	0.0027±0.0012	0.0040±0.0000
Cytb-413(2/5)	0.1073±0.0013	0.1060±0.0000	0.0048±0.0031	0.0058±0.0024	0.0090±0.0000
gnd-396(1/4)	-	-	0.0005±0.0005	0.0010±0.0000	0.0010±0.0000
COI-338(1/4)	-	-	0.0033±0.0021	0.0040±0.0017	0.0060±0.0000
COII-338(1/4)	-	-	0.0027±0.0012	0.0027±0.0012	0.0040±0.0000
Cytb-338(1/4)	-	-	0.0048±0.0031	0.0058±0.0024	0.0090±0.0000
gnd-338(1/4)	-	-	0.0005±0.0005	0.0010±0.0000	0.0010±0.0000

Notes: Interspecific divergences were calculated using the average interspecific distance and smallest interspecific distance. Intraspecific divergences were evaluated by the average intraspecific distance, mean theta, and average coalescent depth. The average interspecific distance was calculated within genera that contained more than one species. The smallest interspecific distance was defined as the minimal value of interspecific distance within genera with at least two species, the average intraspecific distance was the average value of the genetic distances between samples within those species that had at least two individuals, the mean theta was expressed as the average pairwise distance scored from species with more than one obtained representatives by dislodging improper individuals concerned with the asymmetric procurement of samples, and the average coalescent depth was the average value of maximum intraspecific distances.

were (0.0693–0.1233, 0.0060–0.0218), (0.0563–0.1110, 0.0024–0.0070), (0.0703–0.1060, 0.0052–0.0230), and (0.0807–0.1427, 0.0010–0.0090), respectively. The figures above were obtained across the whole dataset. To obtain a more reliable and comparable analysis, we calculated the results of the 338-sample datasets. The smallest interspecific distance and average coalescent depth ranges of COI-338, COII-338, Cytb-338, and gnd-338 were (0.0693–0.0995, 0.0044–0.0165), (0.0563–0.1190, 0.0024–0.0090), (0.0703–0.1040, 0.0052–0.0230), and (0.0811–0.1600, 0.0010–0.0090), respectively. The computations above showed a properly high smallest interspecific distance and a comparatively low average coalescent depth.

To observe the occurrence frequency of different genetic divergences, we drew the frequency line charts of inter- and intra-specific genetic distances based on 338 datasets (Figure 1). Each gene was signified in one chart: the top half was calculated with all

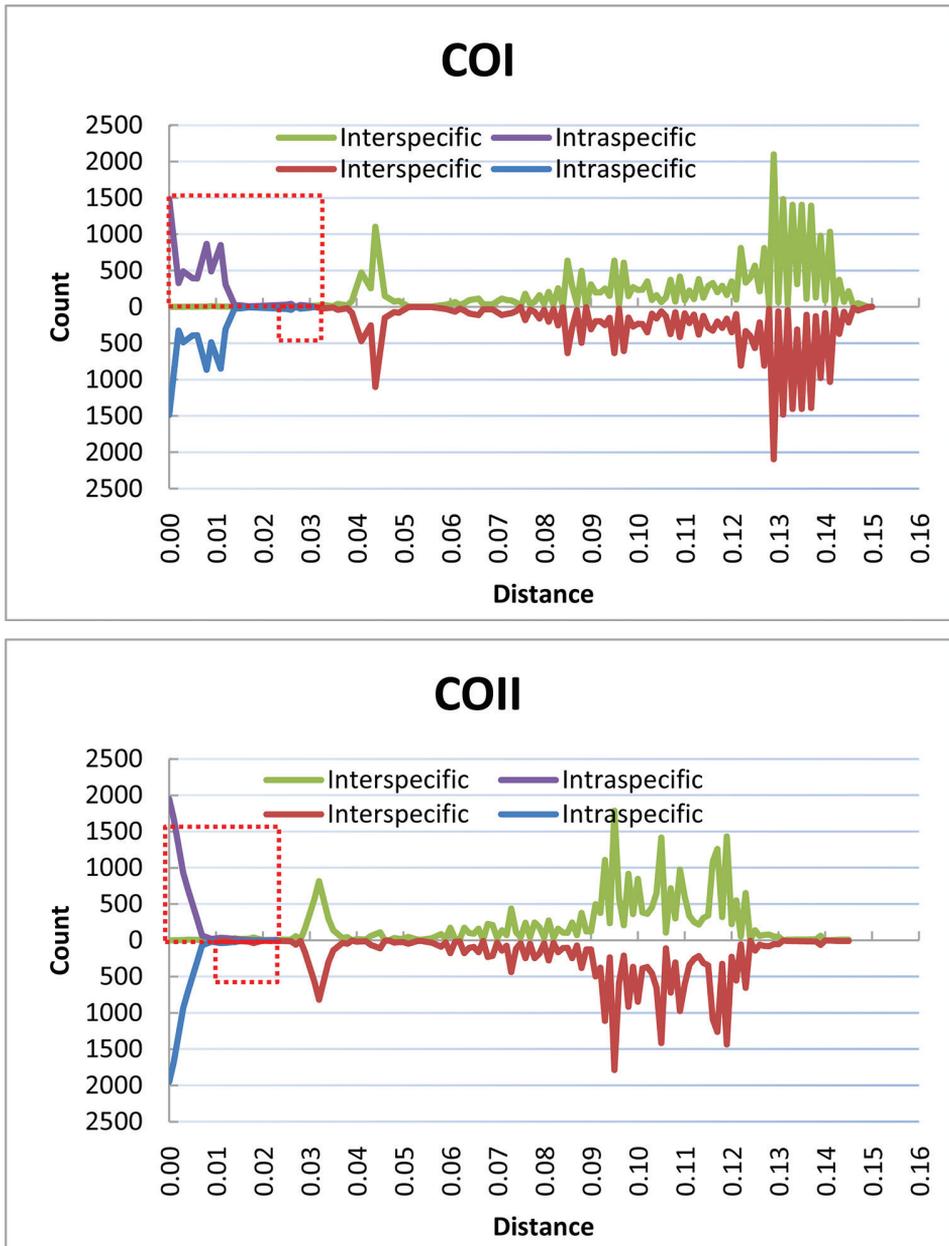


Figure 1. Frequency line charts of inter- and intra-specific genetic distances based on 338 dataset. The x-axis represents the genetic distance, and the y-axis represents the occurrence times in the whole genetic distance matrix. Each peak was a data point with corresponding genetic distance and occurrence times. The data points on the green and red line were calculated with the interspecific distances, and the points on purple and blue line were calculated with the intraspecific distances. The overlap region, which was the crossing area of inter- and intra-specific divergence, is indicated by the red dotted rectangle. Each gene was signified in one chart: the top half was calculated with all the 338 samples; and the bottom half was scored by eliminating the queried samples of *Chaetosiphella longirostris*.

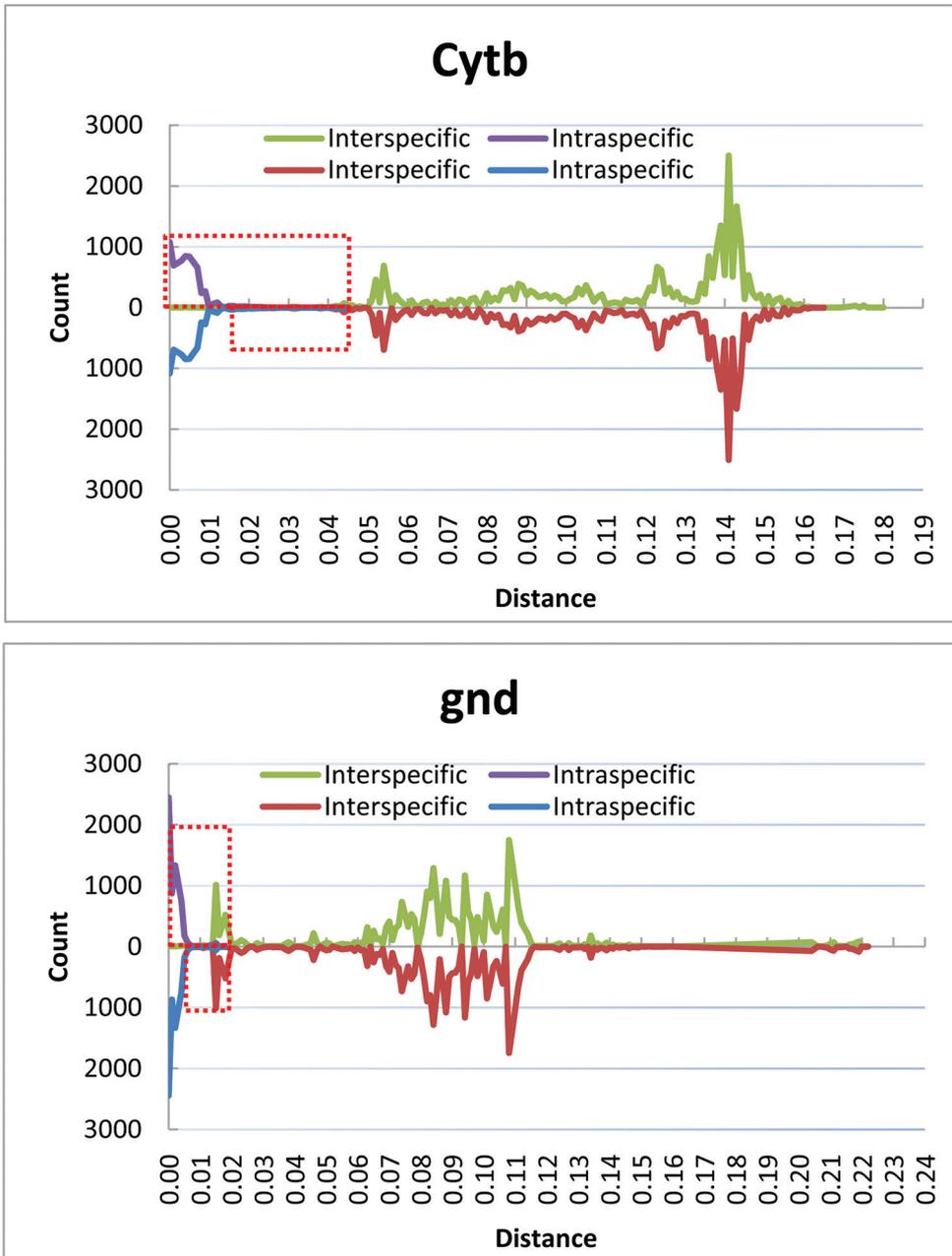


Figure 1. Continue.

338 samples; and the bottom half was scored by eliminating two samples (Nos. 25138 and 25161) of *Chaetosiphella longirostris*. The overlap region was indicated by the red dotted rectangle. No obvious barcoding gap was found in these samples across COI, COII, Cytb, and gnd genes. The overlap regions of COI, COII, Cytb, and gnd in the top half were 0.000–0.031, 0.000–0.023, 0.000–0.045, 0.000–0.018; and those

in the bottom half were 0.022–0.031, 0.011–0.023, 0.015–0.045, 0.006–0.018, respectively. It was clear that the overlap region was much narrower by eliminating the questioned vouchers, and the total frequency within that region was also reduced significantly (Figure 1). The data in the overlap region represented samples that the DNA barcoding would fail to identify. Therefore, a lower total frequency in that region was better. The order of the total frequencies in the overlap region was COI < COII < Cytb < gnd in the top and bottom half parts.

Species delimitation

Given that there were no unambiguous and credible references of genetic thresholds for COII, Cytb, and gnd in aphids, the method of threshold with NJ was applied only in the COI-670 dataset with a threshold of 2% (Footitt et al. 2009b). For all the COI, COII, Cytb and gnd datasets, one distance-based method ABGD (for the analysis results, see Suppl. material 3 for details) and two tree-based approaches GMYC and (b) PTP were employed simultaneously as reference counterpoints to each other.

The morphological and molecular identification results are shown in Table 2. To suitably display the analysis results, four group names were used: *accurate*, of which the putative species clusters were identical with morphological identifications; *split*, whose component vouchers were merely part of a specific unabridged morphological species without representatives of others; *lumped*, which was defined as an aggregation of more than one species including all samples of those species; and partial lumped, a multi-species cluster that consisted of all the vouchers of one or several species as well as part samples of other species.

Seventy-five morphological species were identified from COI-670, which included sequences downloaded from NCBI. For COI-670, we obtained 89 putative species by the GMYC approach with a 65.17% accuracy rate, 85 species using PTP with 67.06% accuracy, 81 species using ABGD with 72.84% accuracy, and 81 species by threshold-NJ with 72.84% accuracy. The COII-376 dataset with only one sequence from NCBI contained 51 morphological species, and 53 hypothetical species were gleaned using the GMYC method with an accuracy rate of 83.02%, 48 species using PTP with 83.33% accuracy, and 50 species using ABGD with 82.00% accuracy. The Cytb-413 data contained 48 morphological species and generated 54 clusters by the GMYC method with an accuracy rate of 79.63%, 49 species using PTP with 81.63% accuracy, and 48 species using ABGD with 79.17% accuracy. There were 49 morphological species within gnd-396, and the putative species found using the GMYC approach was 46 with an accuracy rate of 86.96%, using the PTP method was 45 species with an accuracy rate of 84.44%, and using the ABGD analysis was 48 species with 87.50% accuracy. An analysis of COI-338, COII-338, Cytb-338, and gnd-338 were performed to compare the results of different genes with diverse methods under the same sample composition. There were 45 morphological species within the given 338 samples. The analysis results of various genes and accuracy rate were: COI-338 (GMYC: 47, 89.36%;

Table 2. Analysis results of different datasets with various approaches.

Dataset/Method	Morphology	Cluster number	Accurate	Split	Lumped	Partial lumped
COI-670	75					
GMYC		89	65.17%	31.46%	2.25%	1.12%
PTP		85	67.06%	28.24%	3.53%	1.18%
ABGD		81	72.84%	23.46%	2.47%	1.23%
threshold with NJ		81	72.84%	23.46%	2.47%	1.23%
COII-376	51					1.89%
GMYC		53	83.02%	13.21%	1.89%	
PTP		48	83.33%	8.33%	8.33%	0.00%
ABGD		50	82.00%	12.00%	6.00%	0.00%
Cytb-413	48					
GMYC		54	79.63%	18.52%	0.00%	1.85%
PTP		49	81.63%	12.24%	2.04%	4.08%
ABGD		48	79.17%	12.50%	4.17%	4.17%
gnd-396	49					
GMYC		46	86.96%	4.35%	8.70%	0.00%
PTP		45	84.44%	4.44%	11.11%	0.00%
ABGD		48	87.50%	6.25%	4.17%	2.08%
COI-338	45					
GMYC		47	89.36%	8.51%	0.00%	2.13%
PTP		49	85.71%	12.24%	0.00%	2.04%
ABGD		46	91.30%	6.52%	0.00%	2.17%
COII-338	45					
GMYC		45	93.33%	4.44%	2.22%	0.00%
PTP		45	86.67%	8.89%	4.44%	0.00%
ABGD		45	86.67%	8.89%	4.44%	0.00%
Cytb-338	45					
GMYC		50	80.00%	18.00%	0.00%	2.00%
PTP		46	80.43%	13.04%	2.17%	4.35%
ABGD		42	83.33%	4.76%	7.14%	4.76%
gnd-338	45					
GMYC		42	92.86%	0.00%	7.14%	0.00%
PTP		41	90.24%	0.00%	9.76%	0.00%
ABGD		44	93.18%	2.27%	2.27%	2.27%

Notes: The morphology column gives the number of morphological species of different datasets. Cluster number represents the putative species amount of each method. Accurate represents which putative species clusters were identical with morphological identifications; and split represents which component vouchers were merely part of a specific unabridged morphological species without representatives of others. Lumped, was defined as an aggregation of more than one species and the entire samples of those species were included, whereas partial lumped was defined as a multi-species cluster, which consisted of all the vouchers of one or several species as well as part samples of other species.

PTP: 49, 85.71%; ABGD: 46, 91.30%), COII-338 (GMYC: 45, 93.33%; PTP: 45, 86.67%; ABGD: 45, 86.67%), Cytb-338 (GMYC: 50, 80.00%; PTP: 46, 80.43%; ABGD: 42, 83.33%), and gnd-338 (GMYC: 42, 92.86%; PTP: 41, 90.24%; ABGD:

44, 93.18%). The final results were all displayed in NJ trees (see Suppl. material 4–11) with bootstrap values. The tree topology should be regarded only as distance affinity and not a phylogenetic relationship.

Discussion

The appropriate DNA barcoding and suitable analysis method to Chaitophorinae

COI has not been the only gene marker used for aphid DNA barcoding, other genes from the mitochondrial genome and from endosymbionts having been used for various aphid groups (Chen et al. 2012, Chen et al. 2013a, Liu et al. 2013, Tang et al. 2015). Additionally, many analytical methods have been used in DNA barcoding. Here, we focused on COI, COII, Cytb, and gnd genes, and applied four methods, threshold with NJ, ABGD, GMYC, and PTP, to delimitate species of Chaitophorinae.

For different genes, the amplification efficiency was COI (100%) > Cytb (97%) > gnd (93%) > COII (88%). Within the 338-sample dataset, the difference in values between the smallest interspecific distance and average coalescent depth were unequal in different groups. For *Chaitophorus*, *Periphyllus*, and *Chaetosiphella* (Table 1), the difference values were COI > Cytb > COII > gnd, gnd > COI > COII > Cytb, and gnd > COI > COII = Cytb, respectively. From the overlap region and total frequency (Figure 1), COI and COII were similar with a narrower overlap region and less frequency than Cytb. Although the overlap span of gnd was sufficient, its total frequency in that region was slightly larger. Therefore, the COI and COII genes may be better markers for DNA barcoding.

The most important factor in choosing the delimitation method was the identification accuracy within different genes. Therefore, a better approach means higher identification accuracy and a greater range of application with various genes. The accuracy of GMYC, PTP, and ABGD within COI-338, COII-338, Cytb-338, and gnd-338 were ABGD (91.30%) > GMYC (89.36%) > PTP (85.71%), GMYC (93.33%) > PTP = ABGD (86.67%), ABGD (83.33%) > PTP (80.43%) > GMYC (80.00%), and ABGD (93.18%) > GMYC (92.86%) > PTP (90.24%), respectively (Table 2). In Chaitophorinae, the ABGD was a much better analytical method, and GMYC was also better than PTP in tree-based approaches. Considering that the analysis of ABGD required prior intraspecific distance, a tree-based method needed to be employed concurrently. A tree-based approach should be crosschecked against a non-tree-based approach within species delimitation studies (Fontaneto et al. 2015). As different methods may yield inconformity conclusions (Carstens et al. 2013), the accurate identification of species requires further integrative analysis (Puillandre et al. 2012b, Pante et al. 2015). Herein, a brief investigation of the morphological characteristics combined with a distance-based method of ABGD and a tree-based method of GMYC may be a very suitable pattern for species delimitation and the rapid identification of Chaitophorinae.

DNA barcoding may reveal population differentiation driven by geographical distribution

Chaitophorus saliniger Shinji is an important pest on willows in East Asia. Based on the topology structure and results of analysis with different methods (Figure 2A, Suppl. material 4–11), all samples of this species were divided into two clades which could be regarded as two different species. However, based on the morphological characteristics, all samples should be *Chaitophorus saliniger*. On examining the geographical distribution information of all samples, we found that one of the clades consisted of two samples (Nos. 17651 and 33320) of *C. saliniger* collected from Northeast China, Heilongjiang Province, and the voucher of another sequence (*C. saliniger*-GU978785.1, a downloaded sequence from NCBI) from the Korean Peninsula. The location sites of these three samples were all at a relatively high latitude. All samples in the other clade were not from the aforementioned regions. So, within *C. saliniger*, a population differentiation emerged among the samples from different locations. The genetic divergences between the two clades for COI, Cytb, and gnd were 0.043, 0.039, and 0.020, respectively, which could be regarded as interspecific distances. Although the morphological characteristics of all samples were similar, differentiation at the gene level seems to have occurred between northern and southern populations. Similar genetic differentiation between these populations has also been demonstrated for a nuclear gene, EF-1 α (Fang et al. 2016).

In a similar manner, two samples (Nos. 17613 and 19950) of *Laingia psammae* Theobald were divided into two independent clades (Figure 2B), a result supported at all genes and using different approaches. Sample No. 17613 was from Jilin, northeastern China, whereas sample No. 19950 was from Xinjiang, northwestern China. The genetic distances of three genes (COI, COII and Cytb) between the two samples were 0.064, 0.068, and 0.062, respectively, which reach the level of species (Wang and Qiao 2009). Therefore, differentiation between northeastern and northwestern populations in *L. psammae* exists.

From the topology structures and the constructed consequences of threshold with NJ, ABGD, GMYC, and PTP, we observed that population differentiation was clearly present within both *C. saliniger* and *L. psammae*. Similar findings have been reported in other aphid species (Lee et al. 2011, Wang et al. 2011). The prominent differences among populations may even be an indication of cryptic species (Bickford et al. 2007). Speciation is a long and continuous process, and cryptic species are not easily explained. Within the process, the incipient species may hold for millions of years (Avisé and Walker 1998). Therefore, cryptic species need further study with more samples, combined with morphological characteristics and biological information.

Conclusions

In this work, the DNA barcoding of Chaitophorinae aphids was investigated. Three mitochondrial genes and one endosymbiont gene were used to calculate and compare

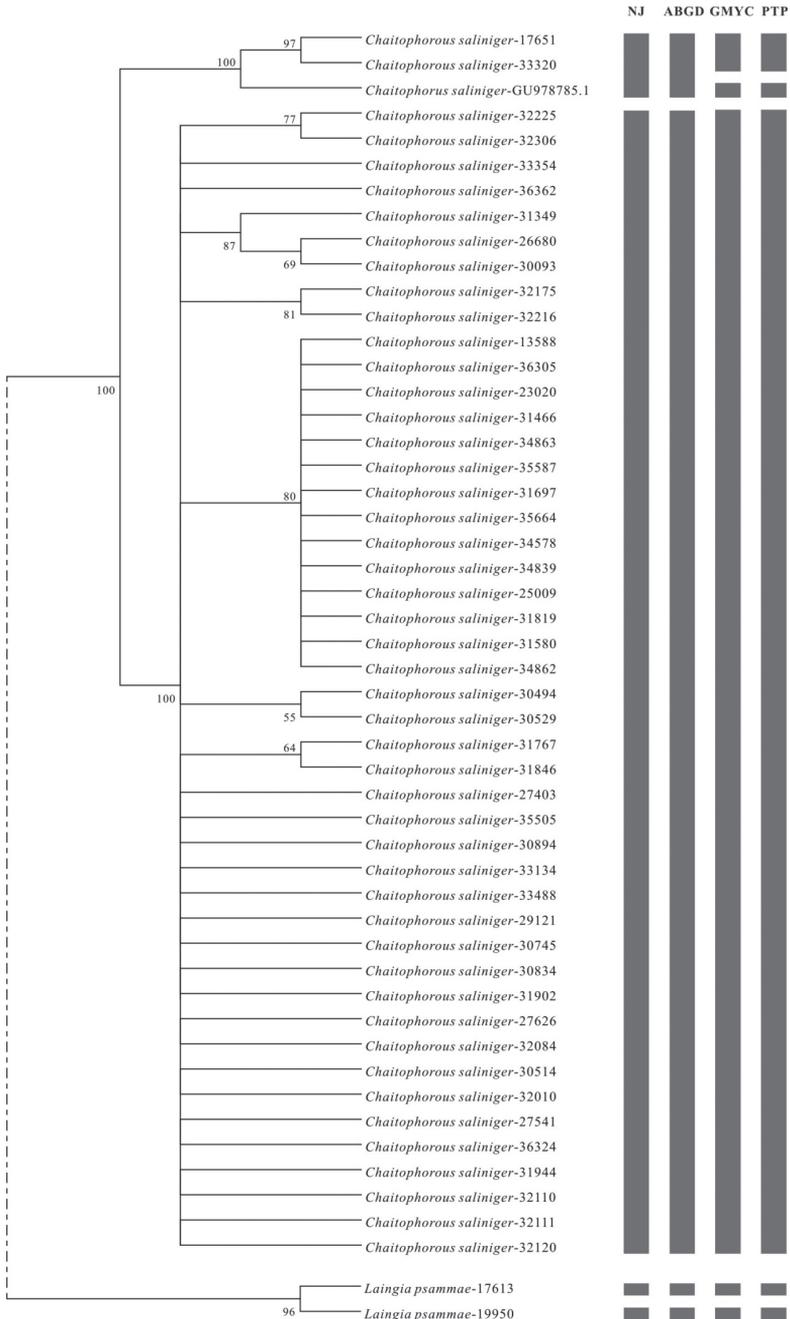


Figure 2. The analysis results of some species from the COI-670 dataset. The analysis results based on other genes were almost identical. The NJ tree was constructed based on the Kimura 2-parameter (K2P) model with a bootstrap value over 50% displayed. The gray blocks behind the tree represent the putative species, which means that the taxa in the tree corresponding to a single block are in one putative species. The number of blocks express the number of putative species using this method. **A** *Chaitophorus saliniger* **B** *Laingia psammae*.

the genetic distances within different datasets. For the delimitation of species, two distance-based methods, threshold with NJ and ABGD, and two tree-based approaches, GMYC and PTP were employed. The interspecific genetic divergence was clearly greater than intraspecific divergence in the four molecular markers. Additionally, the COI and COII genes were more suitable as *Chaitophorinae* DNA barcoding markers. Based on the data for *Chaitophorus saliniger* and *Laingia psammae*, DNA barcoding may reveal population differentiation driven by geographical distribution.

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Supplementary material 1

Table S1

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: specimens data

Explanation note: Sample information.

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Supplementary material 2

Table S2

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: Primer information.

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Supplementary material 3

Table S3

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results with ABGD of all datasets.

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Supplementary material 4

Figure S1

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset COI-670.

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Supplementary material 5

Figure S2

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset COII-376.

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Supplementary material 6

Figure S3

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset Cytb-413.

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Supplementary material 7

Figure S4

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset gnd-396.

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Supplementary material 8

Figure S5

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset COI-338.

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Supplementary material 9

Figure S6

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset COII-338.

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Supplementary material 10

Figure S7

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset Cytb-338.

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Supplementary material 11

Figure S8

Authors: Xi-Chao Zhu, Jing Chen, Rui Chen, Li-Yun Jiang, Ge-Xia Qiao

Data type: molecular data

Explanation note: The analysis results of dataset gnd-338.

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Diversity and distribution of polyphagan water beetles (Coleoptera) in the Lake St Lucia system, South Africa

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Abstract

Water beetles belonging to the suborder Polyphaga vary greatly in larval and adult ecologies, and fulfil important functional roles in shallow-water ecosystems by processing plant material, scavenging and through predation. This study investigates the species richness and composition of aquatic polyphagan assemblages in and around the St Lucia estuarine lake (South Africa), within the iSimangaliso Wetland Park, a UNESCO World Heritage Site. A total of 32 sites were sampled over three consecutive collection trips between 2013 and 2015. The sites encompassed a broad range of aquatic habitats, being representative of the variety of freshwater and estuarine environments present on the St Lucia coastal plain. Thirty-seven polyphagan taxa were recorded during the dedicated surveys of this study, in addition to seven species-level records from historical collections. Most beetles recorded are relatively widespread Afrotropical species and only three are endemic to South Africa. Samples were dominated by members of the Hydrophilidae (27 taxa), one of which was new to science (*Hydrobiomorpha perissinottoi* Bilton, 2016). Despite the fauna being dominated by relatively widespread taxa, five represent new records for South Africa, highlighting the poor state of knowledge on water beetle distribution patterns in the region. Wetlands within the dense woodland characterising the False Bay region of St Lucia supported a distinct assemblage of polyphagan beetles, whilst sites occurring on the Eastern and Western Shores of Lake St Lucia were very similar in their beetle composition. In line with the Afrotropical region as a whole, the aquatic Polyphaga of St Lucia appear to be less diverse than the Hydradephaga, for which 68 species were recorded during the same period. However, the results of the present study, in conjunction with those for Hydradephaga, show that the iSimangaliso Wetland Park contains a high beetle diversity. The ongoing and future ecological protection of not only the estuarine lake itself, but also surrounding freshwater wetlands, is imperative and should be taken into consideration during future management planning for the park.

Keywords

Afrotropical region, biodiversity census, aquatic Coleoptera, Polyphaga, Hydrophilidae, iSimangaliso Wetland Park

Introduction

A recent survey of the Hydradephaga of the Lake St Lucia system, located within the iSimangaliso Wetland Park, KwaZulu-Natal, South Africa, has shown that this is a hot-spot of aquatic beetle diversity, with 68 species recorded in total, including several new records for the region (Perissinotto et al. 2016). This paper details the polyphagan water beetles found during the course of the same surveys.

The suborder Polyphaga includes the vast majority of beetles, with an estimated 320 000 species currently described in 151 families (Beutel and Leschen 2005). On a global scale, the diversity of Polyphaga that inhabit truly aquatic environments is slightly above that of the Hydradephaga, although in the Afrotropical region more Adephaga have been described to date (Jäch and Balke 2008). Worldwide, thirteen families of Polyphaga have predominantly aquatic representatives, with the Hydrophilidae, Hydraenidae, Scirtidae and Elmidae comprising the bulk of them (Jäch and Balke 2008). Other predominant aquatic families occurring in southern Africa within the suborder are Helophoridae (largely confined to the Palearctic Region, with only two species of *Helophorus* Fabricius, 1775, known from Africa), Epimetopidae (represented in Africa by only one genus, *Eupotemus* Ji & Jach, 1998), Heteroceridae (23 known southern African species), Hydrochidae (with eleven species of *Hydrochus* Leach, 1817, represented in southern Africa), Ptilodactylidae (in a state of taxonomic disarray, but with at least two genera in southern Africa), Spercheidae (monogeneric family with five species recognized in southern Africa), Dryopidae (represented in southern Africa by three genera and eight species); and Psephenidae (with three monospecific genera currently recognized in southern Africa) (Stals and de Moor 2007). Other polyphagans that have affinity for aquatic habitats but are not regarded as true water beetles are now referred to as “paraquatic” (sensu Jäch and Balke 2008). These include the “shore beetles”, “facultative water beetles” and “parasitic water beetles” (sensu Jäch 1998).

Like the hydradephagans, polyphagans are also found in all types of aquatic habitats and although they do not spread into the open ocean, some species are able to tolerate hypersaline conditions as high as 250‰, especially hydraenids in the genus *Ochthebius* Leach, 1815 (Perkins 1980, Gerdes et al. 1985, Abellán et al. 2007). Most species of aquatic Polyphaga are either scavengers or phytophages, but some of the larger species are predatory, particularly in the Hydrophiloidea, which also frequently have aquatic larvae, some of which may be semi-terrestrial (Beutel and Leschen 2005). Thus they play key roles in aquatic ecosystems and may significantly impact the trophic structure and functioning of wetland ecosystems, such as Lake St Lucia and its associated wetlands in northern KwaZulu-Natal, South Africa.

The St Lucia lake system is part of the iSimangaliso Wetland Park, South Africa's first UNESCO World Heritage Site and a RAMSAR Wetland of International Importance (Perissinotto et al. 2013). The broad region of the park has historically experienced drastic shifts in climatic conditions, with droughts and floods alternating at semi-decadal cycles (Perissinotto et al. 2013). The last wet phase was recorded in the park from 2011 to 2014, resulting in repeated flood events, with large amounts of freshwater flowing into the system, changing the prevailing salinity state from hypersaline to oligo- or polyhaline. This led to the penetration of numerous brackish and freshwater species into the estuarine lake from adjacent rivers and wetlands. Prominent among these were aquatic insects, especially beetles, thereby providing an opportunity and necessity to renew efforts towards the investigation of the diversity and dynamics of this invertebrate component within the system. Here we report the findings of a census on the aquatic Polyphaga undertaken within Lake St Lucia and its associated wetlands during the period July 2013–February 2015. The results of this have been combined with historical records to provide a baseline for future identification and monitoring of beetle biodiversity patterns in response to climatic and anthropogenic changes.

Materials and methods

The sampling design and protocol for this study follow those described by Perissinotto et al. (2016), who provide a detailed description of study sites and sampling techniques. A summary of their account is provided here.

Study area

Lake St Lucia (27°52'0"S to 28°24'0"S and 32°21'0"E to 32°34'0"E) is located in the north-eastern corner of South Africa in the KwaZulu-Natal province and is a large (~ 300 to 350 km²) estuarine lake system comprising three interconnected shallow lakes (South Lake, North Lake and False Bay) that are joined to the Indian Ocean via a 21 km channel known as the Narrows (Fig. 1). Aquatic beetle samples were collected from a variety of freshwater habitats on the coastal plain surrounding the main expanse of Lake St Lucia, and from the vegetated margins of the estuarine lake body itself. Three collection trips were undertaken: November 2013 (19th–30th); July 2014 (23rd–24th); and January–February 2015 (31st January – 6th February). In total, 32 sites were sampled over the course of the three collection trips, encompassing a diverse array of habitats (Fig. 1).

Six waterbody types were sampled (following the classification of Ollis et al. 2015): depression wetlands (both isolated and non-isolated, n = 16); valley bottom wetlands (both channelled and un-channelled, n = 8); rivers (both in-channel and riparian habitats, n = 4); a wetland flat; a seepage wetland; and the estuarine lake body itself. Most

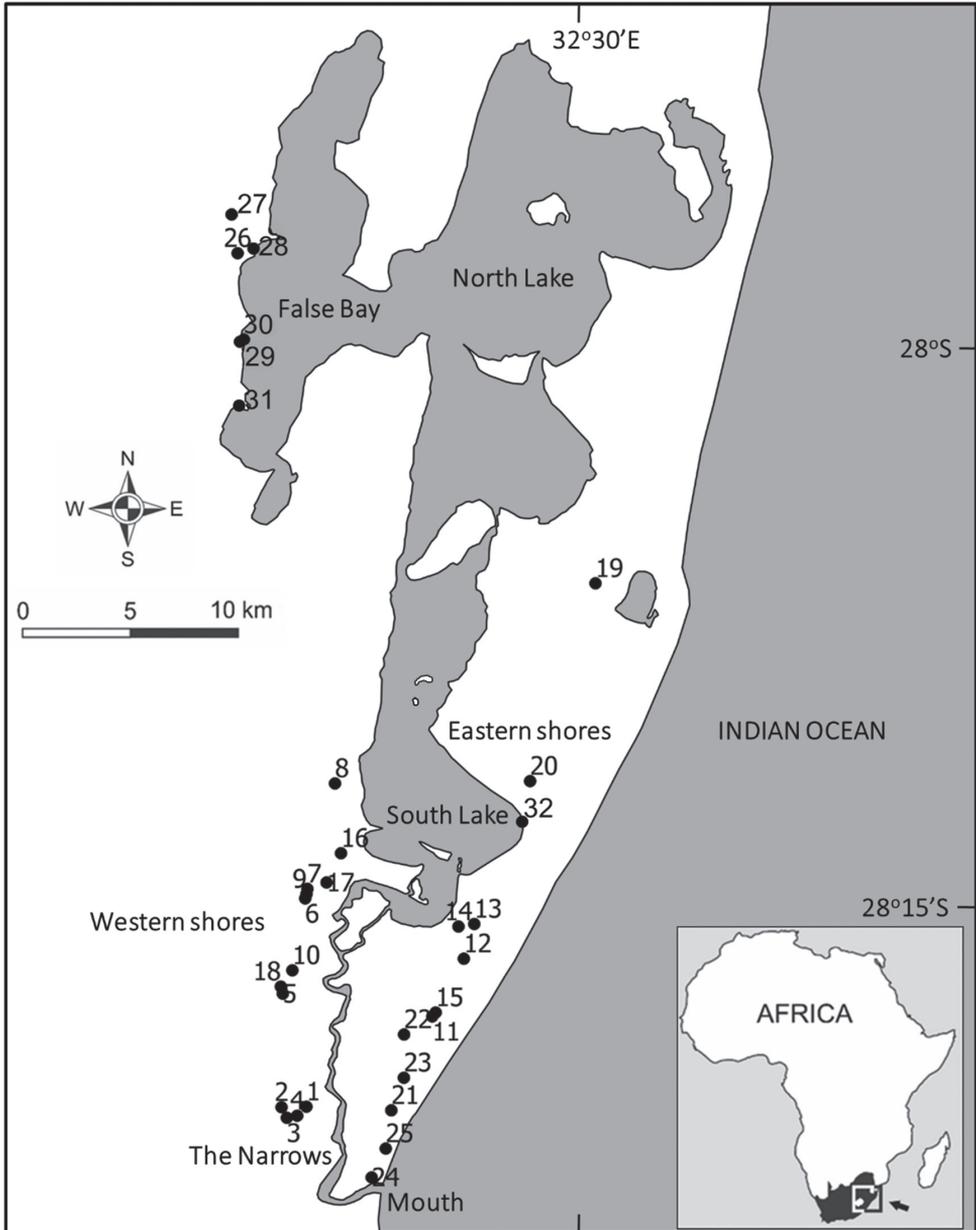


Figure 1. The Lake St Lucia system in northern KwaZulu-Natal. The locations of sites sampled between November 2013 and February 2015 are depicted. Site numbers 1–32 correspond to those in Table 1. Figure reproduced with permission from Perissinotto et al. (2016).

of these waterbodies were extensively vegetated and some of the smaller depression and valley bottom wetlands were temporary. The locations sampled, their habitat classification and dates of sampling are summarised in Table 1.

Table 1. Geographic position and classification of the waterbodies sampled during this study. Sampling took place during the three collecting trips to Lake St Lucia during November 2013, July 2014 and January/February 2015. Classification (wetland type) follows the hydrogeomorphic (HGM) approach of Ollis et al. (2015). WS – Western Shores; ES – Eastern Shores; FB – False Bay. Table reproduced with permission from Perissinotto et al. (2016).

Site	GPS (D°M'S")		Wetland type	Region	Nov 2013	Jul 2014	Jan/ Feb 2015
1	28°20'53.33"S	32°23'38.42"E	River (pool)	WS		×	×
2	28°20'54.23"S	32°22'59.68"E	Depression	WS		×	
3	28°21'10.77"S	32°23'7.88"E	Channelled valley bottom	WS		×	
4	28°21'7.52"S	32°23'24.04"E	Channelled valley bottom	WS		×	
5	28°17'55.76"S	32°23'10.62"E	River (riparian zone)	WS		×	
6	28°15'26.06"S	32°23'36.51"E	Depression	WS	×	×	×
7	28°15'11.10"S	32°23'39.95"E	Depression	WS	×	×	×
8	28°12'25.44"S	32°24'22.97"E	Depression (artificial)	WS		×	
9	28°15'19.19"S	32°23'38.53"E	Depression	WS		×	
10	28°17'19.08"S	32°23'16.53"E	Depression	WS		×	
11	28°18'31.52"S	32°26'54.54"E	Un-channelled valley bottom	ES		×	
12	28°17'00.81"S	32°27'43.78"E	Depression	ES		×	
13	28°16'6.26"S	32°28'00.02"E	Depression	ES		×	×
14	28°16'10.26"S	32°27'35.43"E	Depression	ES		×	×
15	28°18'25.29"S	32°26'59.88"E	Un-channelled valley bottom	ES		×	
16	28°14'15.05"S	32°24'32.30"E	Depression	WS			×
17	28°15'1.00"S	32°24'9.85"E	Channelled valley bottom	WS			×
18	28°17'44.59"S	32°22'58.49"E	Flat	WS			×
19	28°07'10.99"S	32°31'8.98"E	Un-channelled valley bottom	ES			×
20	28°12'21.75"S	32°29'27.07"E	River (main channel)	ES			×
21	28°20'59.06"S	32°25'50.76"E	Depression	ES			×
22	28°18'59.92"S	32°26'10.64"E	Depression	ES			×
23	28°20'7.84"S	32°26'10.36"E	Depression	ES			×
24	28°22'44.46"S	32°25'20.13"E	River (connected to estuary)	ES			×
25	28°21'59.12"S	32°25'42.10"E	Depression	ES			×
26	27°58'32.33"S	32°21'51.14"E	Depression	FB			×
27	27°57'31.50"S	32°21'41.82"E	Depression	FB	×		×
28	27°58'25.01"S	32°22'16.02"E	Channelled valley bottom	FB			×
29	28°00'51.44"S	32°21'54.93"E	Channelled valley bottom	FB	×		×
30	28°00'47.95"S	32°22'00.92"E	Estuarine lake	FB	×		×
31	28°02'9.17"S	32°21'42.78"E	Estuarine lake shore (light trap)	FB	×	×	×
32	28°13'14.56"S	32°29'12.45"E	Seep	ES			×

Field sampling protocol

Beetle collection efforts primarily involved the use of a long-handled square-framed sweep net (30 cm mouth and 1 mm mesh), following a sweep protocol similar to that of Bilton et al. (2006) and Bird et al. (2013). Sampling effort was concentrated in

submerged vegetation and at the shore margins. A semi-quantitative approach was incorporated, whereby approximately 20 sweeps from the water surface to bottom substrate and back to the surface were performed at each waterbody. Visual searches for beetles at the shore margin and light trapping were also conducted to complement the array of taxa collected with the sweep net. The light trap consisted of a 4×3 m white sheet that was hung vertically below a fluorescent mercury vapour lamp (Radiant 250 W) and was deployed on all three survey trips at False Bay, adjacent to the lake shore (site 31, Table 1), during the evening (20:00–05:00 hrs). Aquatic coleopteran specimens were retrieved from the light sheet by hand. All beetle specimens collected during the November 2013 and July 2014 surveys were killed using ethyl acetate vapour and preserved in 5% formalin solution. Specimens collected during January–February 2015 were killed in the same way and preserved in 70% ethanol.

A range of *in situ* physico-chemical parameters were measured at each site. Salinity, temperature, pH, dissolved oxygen and turbidity were recorded using a YSI 6600-V2 multi-system probe. Due to technical problems, physico-chemical measurements were not taken during November 2013.

Historical data and other collections

Aquatic Polyphaga collections housed in the major South African museums, namely the Iziko South African Museum (ISAM, Cape Town), the Ditsong National Museum of Natural History (DNMNH, Pretoria; formerly the Transvaal Museum) and the South African National Collection of Insects (SANC, Pretoria), were databased by the respective curators to add historical records to this study. Further data on species collected during previous surveys in the St Lucia area were obtained from Day et al. (1954), Millard and Broekhuysen (1970), Vrdoljak (2004) and Perkins (2014). Records of specimens collected by the authors of the current study during preliminary investigations in the area carried out between 2008 and 2012 were also included. Most of this historical material has, however, not been examined by taxonomic specialists, except for the 2008–2012 collections. Identifications should therefore be considered with caution.

Identification and illustrations

Species identification was undertaken with reference to museum material and the most recent literature available on the specific taxa. Characteristics of male genitalia were generally used as the key criterion for species identification and separation. Digital photographs of the dorsal habitus of each species were taken using a EOS 600D digital camera fitted to a Sigma 50mm f/2.8 EX DG macro lens for larger specimens (≥ 1.5 cm) and a Leica Z6 APO for smaller specimens (< 1.5 cm). Image stacks were

produced by hand, and combined using Zerene Stacker software (www.zerenesystems.com). To facilitate future identification and monitoring exercises, an annotated and illustrated list was compiled of all species identified in the preliminary collections of 2008-2012 and during the three dedicated surveys conducted in November 2013, July 2014 and January/February 2015 (Appendix 1).

Statistical analysis

Multivariate techniques were used to analyse spatial trends in the composition of polyphagan beetle assemblages at St Lucia. Beetle data were converted to presence-absence and assemblage similarity amongst sites was analysed using the Bray-Curtis coefficient. Non-metric multidimensional scaling (MDS) was used to depict beetle assemblages at St Lucia on a two-dimensional plot. Differences in beetle assemblage composition across the regions of St Lucia (Eastern Shores, Western Shores and False Bay) and waterbody types (excluding seeps and flats as only one of each was sampled) were tested using non-parametric permutational MANOVA (PERMANOVA, Anderson 2001), using the Bray-Curtis coefficient for construction of the dissimilarity matrix. As with the multivariate data (assemblage composition), the univariate measure of species richness (number of species per waterbody) was also compared across both regions and waterbody types, this time using the non-parametric Kruskal-Wallis test. Linear regression was used to assess the relationship between the number of polyphagan taxa recorded per site and that recorded for the adephagans by Perissinotto et al. (2016).

Multidimensional scaling was performed using PRIMER v6 software (Clarke and Warwick 2001, Clarke and Gorley 2006). Non-parametric permutational MANOVA was performed using the PERMANOVA routine in the PERMANOVA+ add-on package (Anderson et al. 2008) to PRIMER v6 software. P-values for PERMANOVA models were tested using 9999 unrestricted permutations of the raw data. The Kruskal-Wallis tests on species richness and linear regression were performed using Statistica 12 software for Windows (Statsoft Inc. 2015). All tests were performed using an *a priori* significance level of $\alpha = 0.05$.

Results

The sites sampled during this survey reflect the relative abundance of the various waterbody types encountered on the St Lucia coastal plain, with groundwater-fed depressions and valley bottom wetlands predominating, although several small rivers, a wetland flat and a seep were also sampled, in addition to the estuarine lake itself. Freshwater wetlands around Lake St Lucia were mostly small (< 2 ha), shallow (< 1 m maximum depth) and extensively vegetated. Further details on the physico-chemistry of the waterbodies sampled at St Lucia are provided by Perissinotto et al. (2016).

Polyphaga collected during the current survey

A total of 37 taxa of aquatic Polyphaga were collected during the three dedicated surveys of the current study (2013–2015), which are listed in Table 2 and illustrated in the checklist (see Appendix 1). The survey revealed a new species of Hydrophilidae, recently described as *Hydrobiomorpha perissinottoi* Bilton, 2016 (Bilton 2016). In addition, five species represent new records for the Republic of South Africa, four of which are hydrophilids (*Paracymus exiguus* Wooldridge, 1977; *Amphiops uhligi* Hebauer, 1995; *Hydrochara fulvofemorata* (Fairmaire, 1869) and *Laccobius uhligi* Gentili, 1995). The other species is a hydraenid (*Aulachochthebius* cf. *continentalis* (Orchymont, 1929)), which, whilst new to South Africa, has not been identified with certainty. This genus is currently being revised (Phil Perkins, pers. comm.). Of the 37 polyphagan taxa collected in this study, 27 were identified to species level. The remaining 10 taxa were assigned to the following (sub)genera, and could not be named reliably to species: *Hydrochus*; *Allocotocerus* Kraatz, 1883; *Enochrus* (*Methydrus*) Thomson, 1859; *Helochares* Mulsant, 1844; and *Coelostoma* Brullé, 1835. In the case of these genera, taxa were assigned to morphospecies, but further taxonomic work, including in some cases generic revisions, would be required to name species with confidence, but such taxonomic uncertainty does not affect our analyses. Hydrophilidae dominated the polyphagan beetle assemblages at St Lucia, being represented by 27 species. Relatively minor representation was afforded by the Hydrochidae (three species of *Hydrochus*); Spercheidae (two species, *Spercheus cerisyi* Guérin-Ménéville, 1842 and *S. senegalensis* Castelnau, 1832); Hydraenidae (four species, *Hydraena cooperi* Balfour-Browne, 1954, *Limnebius probus* Perkins, 2015, *Aulachochthebius* cf. *continentalis* and *Ochthebius andronicus* Orchymont, 1948); and Curculionidae (one species, *Pseudobagous* cf. *longulus* (Gyllenhal, 1836)).

Polyphagan beetles were generally widespread across a number of waterbodies, with 21 of the 37 species being collected from five or more sites (Table 2). The three most widespread species were *Helochares* sp. 2, collected from 18 waterbodies, and *Hydrochus* sp. 1 and *Enochrus* (*Methydrus*) sp., both collected from 16 waterbodies. Only five species (*Hydrochus* sp. 3, *Paracymus amplus*, *Amphiops uhligi*, *Hydrochara elliptica* and *Laccobius uhligi*) were recorded from a single waterbody (Table 2).

Historical records

Polyphagan taxa collected from St Lucia and surroundings prior to the current survey are listed in Table 3. A total of 49 taxa were previously recorded from the region, but of these, only 19 represent species-level records. Nineteen unpublished museum records were found from our extensive search of museum collection records across South Africa (including material stored by the authors at UKZN). Of these, 15 are species-level records, 10 of which were also recorded during the collections of the current study (2013–2015: indicated by an asterisk in Table 3). Thirty-one aquatic polyphagan taxa

Table 2. Polyphagan beetles collected from St Lucia during the course of this study. The sites are listed from which each taxon was collected on each of the three sampling trips. Site numbers 1 – 32 correspond to those listed in Table 1. The regions where each taxon occurred are also indicated: WS – Western Shores; ES – Eastern Shores; FB – False Bay. Species new to South Africa are shown in bold type. Classification of Hydrophilidae follows Short and Fikáček (2013).

Taxon	Sampling date			Region		
	Nov 2013	Jul 2014	Jan/Feb 2015	WS	ES	FB
Hydrochidae:						
<i>Hydrochus</i> sp. 1		2, 3, 5, 13, 14, 15	6, 7, 13, 14, 16, 17, 18, 21, 22, 23, 27, 32	×	×	×
<i>Hydrochus</i> sp. 2			6, 14, 17, 19, 27, 32	×	×	×
<i>Hydrochus</i> sp. 3			18	×		
Spercheidae:						
<i>Spercheus cerisyi</i> Guérin-Méneville, 1842			6, 7, 14, 16, 17, 18, 21, 27	×	×	×
<i>Spercheus senegalensis</i> Castelnau, 1832			6, 14, 17, 18, 20, 25, 27	×	×	×
Hydrophilidae:						
<i>Amphiops globus</i> Erichson, 1843			1, 20, 27	×	×	×
<i>Amphiops senegalensis</i> (Laporte, 1840)		15	1, 7, 14, 16, 22, 23, 25, 27	×	×	×
<i>Amphiops uhligi</i> Hebauer, 1995			14		×	
<i>Allocotocerus</i> sp.	27	10	6, 14, 18, 23, 27, 29	×	×	×
<i>Berosus cuspidatus</i> Erichson, 1843			6, 7, 14, 18, 21, 22, 27, 28, 29, 31, 32	×	×	×
<i>Berosus viticollis</i> Boheman, 1851	7, 29, 30			×		×
<i>Regimbartia nilotica</i> (Sharp, 1903)	27		6, 14, 18, 21, 27, 29	×	×	×
<i>Regimbartia obsoleta</i> (Régimbart, 1906)			14, 18, 22, 27, 29	×	×	×
<i>Laccobius uhligi</i> Gentili, 1995			32		×	
+ <i>Paracymus amplus</i> Wooldridge, 1977			21		×	
<i>Paracymus exiguus</i> Wooldridge, 1977			7, 13, 18, 21, 29	×	×	×
<i>Paracymus pisanus</i> Balfour-Browne, 1954			7, 13, 14, 18, 21, 25, 27, 29, 32	×	×	×
*+ <i>Hydrobiomorpha perisinottoi</i> Bilton, 2016			16, 18, 22, 29	×	×	×
<i>Hydrochara elliptica</i> (Fabricius, 1801)	31					×
<i>Hydrochara fulvofemorata</i> (Fairmaire, 1869)	30		6, 16, 17, 26, 27, 29, 31	×		×
<i>Hydrophilus aculeatus</i> (Solier, 1834)	31	10	14, 31	×	×	×
<i>Sternolophus solieri</i> Laporte, 1840	30		14, 17, 18, 20, 22, 23, 24, 27, 31	×	×	×
<i>Enochrus</i> (<i>Methydus</i>) sp.	30	1, 4, 5	7, 13, 14, 16, 17, 18, 19, 21, 27, 28, 29, 32	×	×	×
<i>Chasmogenus</i> cf. <i>patrizii</i> (Balfour-Browne, 1948)			14, 23, 27, 29		×	×
<i>Helochares dilutus</i> (Erichson, 1843)		10	6, 21, 24, 27, 28, 29, 31	×	×	×
<i>Helochares longipalpis</i> (Murray, 1859)		11	14, 16, 17, 22, 23, 27, 29, 31, 32	×	×	×
<i>Helochares</i> sp. 1		12	27		×	×
<i>Helochares</i> sp. 2		3, 4, 5, 15	1, 6, 7, 14, 17, 18, 19, 21, 23, 24, 25, 27, 29, 31	×	×	×
<i>Coelostoma</i> sp. 1			14, 20, 23, 27, 31, 32		×	×

Taxon	Sampling date			Region		
	Nov 2013	Jul 2014	Jan/Feb 2015	WS	ES	FB
<i>Coelostoma</i> sp. 2	29, 30	1, 10	18, 23, 24, 26, 27, 28, 31, 32	×	×	×
<i>Coelostoma</i> sp. 3			22, 23		×	
<i>Cercyon dieganus</i> Régimbart, 1903			1, 14, 16, 22, 23, 27, 28, 31	×	×	×
Hydraenidae:						
<i>Hydraena cooperi</i> Balfour-Browne, 1954			14, 17, 21, 22, 25, 29, 32	×	×	×
+ <i>Limnebius probus</i> Perkins, 2015			27, 29			×
<i>Aulachochthebius</i> cf. <i>continentalis</i> (Orchymont, 1929)			21, 27, 29, 32		×	×
<i>Ochthebius andronicus</i> Orchymont, 1948			21, 29		×	×
Curculionidae:						
<i>Pseudobagous</i> cf. <i>longulus</i> (Gyllenhal, 1836)		13, 14	29		×	×

+ Taxa known only from South Africa.

* New species, first found in this study.

Table 3. Aquatic polyphagan beetles previously recorded from the Lake St Lucia system and surrounding waterbodies. Literature sources are indicated by letters as follows: (a) Day et al. (1954); (b) Millard and Broekhuysen (1970); (c) Vrdoljak (2004); and (d) Perkins (2014). Museum and national collection material is as follows: SANC – South African National Collection of Insects; ISAM – Iziko South African Museum; DNMNH – Ditsong National Museum of Natural History. Locations referred to are: FWW – fresh water wetlands on the Eastern Shores of Lake St Lucia; FB – False Bay; SL – St Lucia (lake body and immediate surrounds); KB – Kosi Bay; D; Dukuduku; DF – Dukuduku forest; DP – Dukandlovu Pan (site 29 in the current study). Also included here are records based on ad hoc collections undertaken by the authors during the period 2008–2012 (deposited at the University of KwaZulu-Natal and listed as UKZN).

Family	Genus	Species	Publication	Years recorded	Location
Hydrochidae	<i>Hydrochus</i> Leach, 1817	<i>Hydrochus</i> spp. 1–4	(c)	2002/2003	FWW
Spercheidae	<i>Spercheus</i> Illiger, 1798	<i>S. cerisyi</i> *	SANC	Not specified	D
		<i>S. senegalensis</i> *	SANC	Not specified	SL, D
Hydrophilidae	<i>Allocotocerus</i> Kraatz, 1883	<i>Allocotocerus</i> sp.	(c)	2002/2003	FWW
	<i>Amphiops</i> Erichson, 1843	<i>A. senegalensis</i> *	(c)	2002/2003	FWW
		<i>Amphiops</i> spp. 1–2	(c)	2002/2003	FWW
	<i>Anacaena</i> Thomson, 1859	<i>Anacaena</i> sp.	(c)	2002/2003	FWW
	<i>Berosus</i> Leach, 1817	<i>B. cuspidatus</i> *	(a), (b)	1948	FB
		<i>Berosus</i> spp. 1–3	DNMNH	1960	SL
	<i>C. rufitarse</i> (Boheman, 1851)	<i>C. rufitarse</i>	(a), (b)	1948	FB
		<i>Coelostoma</i> spp. 1–4	(c)	2002/2003	FWW
	<i>Dactylosternum</i> Wollaston, 1854	<i>Dactylosternum</i> sp.	(c)	2002/2003	FWW
	<i>Enochrus</i> Thomson, 1859	<i>Enochrus</i> spp. 1–4	(c)	2002/2003	FWW
	<i>Helochares</i> Mulsant, 1844	<i>H. dilutus</i> *	SANC	Not specified	SL, D
		<i>H. longipalpis</i> *	SANC	Not specified	SL, D
		<i>Helochares</i> spp. 1–4	(c)	2002/2003	FWW
	<i>Hydrochara</i> Berthold, 1827	<i>H. elliptica</i> *	UKZN	2012	DP
<i>Regimbartia</i> Zaitzev, 1908	<i>Regimbartia</i> sp.	(c)	2002/2003	FWW	

Family	Genus	Species	Publication	Years recorded	Location
Hydrophilidae	<i>Sternolophus</i> Solier, 1834	<i>S. solieri</i> *	UKZN	2008	FB
		<i>S. angolensis</i> (Erichson, 1843)	(c)	2002/2003	FWW
		<i>Sternolophus</i> sp.	(c)	2002/2003	FWW
	<i>Hydrophilus</i> Geoffroy, 1762	<i>H. aculeatus</i> *	SANC	Not specified	D, SL
Hydrophilidae	<i>Hydrophilus</i> Geoffroy, 1762	<i>H. senegalensis</i> (Percheron 1835)	UKZN	2012	FB
		<i>Hydrophilus</i> sp.	ISAM	1988	KB
	<i>Chasmogenus</i> Sharp, 1882	<i>C. lycetus</i> (d'Orchymont, 1939)	SANC	Not specified	SL
		<i>C. patrizii</i> *	SANC	Not specified	SL
Hydraenidae	<i>Hydraena</i> Kugelann, 1794	<i>H. cooperi</i> *	(d)	1997	
Scirtidae	<i>Cyphon</i> Paykull, 1799	<i>Cyphon</i> sp.	DNMNH	Not specified	SL, DF, KB
	<i>Scirtes</i> Illiger, 1807	<i>Scirtes</i> sp.	DNMNH	Not specified	SL, KB
	<i>Ora</i> Clark, 1865	<i>Ora</i> sp.	DNMNH	Not specified	SL
Heteroceridae	<i>Augyles</i> Schiödte, 1866	<i>A. pallens</i> (Charpentier, 1965)	SANC	Not specified	D
	<i>Heterocerus</i> Fabricius, 1792	<i>H. atroincertus</i> Charpentier, 1965	SANC	Not specified	SL
		<i>H. thebaicus australis</i> Charpentier, 1965	SANC	Not specified	D
Curculionidae	<i>Pseudobagous</i> Sharp, 1917	<i>P. longulus</i> *	SANC	Not specified	D

* Also recorded during the dedicated surveys of 2013–2015.

have been reported for St Lucia from previously published studies of the system (unpublished M.Sc. thesis in the case of Vrdoljak 2004), although only five of these are species-level records (three of which were also recorded in the current study), reflecting the ecological rather than taxonomic focus of these studies. Although Heteroceridae and Scirtidae are listed in Table 3, members of these families were not recorded in the collections of the current study.

Patterns of assemblage composition

The composition of polyphagan beetle assemblages was similar between the Western and Eastern Shores of St Lucia, as reflected by the high degree of overlap of sites from these two regions in the MDS plot (Fig. 2a). Assemblages from False Bay show some distinction from those of the other two regions, with sites generally placed toward the left of the plot in Fig. 2a. The PERMANOVA results (Table 4) confirm these patterns, reporting a significant overall difference in assemblage composition of sites across the three regions sampled, with post hoc pairwise tests indicating that the overall difference was driven by the distinctness of the False Bay sites (Table 4a). The different waterbody types at St Lucia did not appear to harbour distinct assemblages of polyphagan beetles, with sites from the different waterbody types overlapping

Table 4. Non-parametric permutational MANOVA (PERMANOVA) results for models comparing beetle assemblage composition. Assemblage composition at St Lucia was compared across (a) regions, and (b) waterbody types. The multivariate models tested for differences between group centroids in Bray-Curtis dissimilarity space. Pairwise comparisons are reported in the case of (a), where overall test results were significant. WS – Western Shores; FB – False Bay; ES – Eastern Shores.

(a)						<i>Post hoc pairwise comparisons</i>		
Source	df	SS	MS	F	P	Groups	t	P
Region	2	13006	6502.9	1.9978	0.012*	WS, FB	1.5753	0.019*
Residual	30	100910	3255.1			WS, ES	0.85389	0.689
Total	32	113910				FB, ES	1.7283	0.002*
(b)								
Source	df	SS	MS	F	P			
Waterbody type	3	13102	4367.4	1.2997	0.144			
Residual	29	100810	3360.4					
Total	32	113910						

* Significant P values at $\alpha = 0.05$.

widely in the MDS plot (Fig. 2b). This overlap is confirmed by the PERMANOVA results, reporting no significant difference in assemblage composition across wetland types (Table 4b).

Species richness patterns

Kruskal-Wallis tests showed that polyphagan beetle richness did not differ significantly between the three regions of St Lucia ($KW-H_{2,37} = 0.9006$, $p = 0.6374$) or between waterbody types ($KW-H_{5,37} = 4.2675$, $p = 0.5116$). Mean richness across all sites and sampling trips was 6.4 ± 5.9 (SD) species per site, the high standard deviation reflecting large variation in the number of species recorded per site. The boxplots in Fig. 3 provide a visual depiction of the median and range of richness values across sites, grouped firstly by region (Fig. 3a) and secondly by waterbody type (Fig. 3b). Although median richness per site was low (~ 4 species) in each of the three regions of St Lucia, the distribution was skewed; the non-outlier ranges of all three regions including sites with 15 or more species (Fig. 3a). In terms of waterbody types, the boxplot (Fig. 3b) does not reveal any clear pattern of differences among groups. ‘Wetland flat’ and ‘seep’ categories were each represented by only a single site sampled on one occasion (January/February 2015) and were both relatively rich in taxa (15 and 12 species). Four sites yielded only a single species (sites 7A, 2B, 11B and 12B; Fig. 3c). The three richest samples were all collected during the mid-summer wet season sampling trip in January/February 2015, from a depression wetland at False Bay (site 27C with 24 species), a depression wetland on the Eastern Shores (site 14C with 20 species) and a valley bottom wetland at

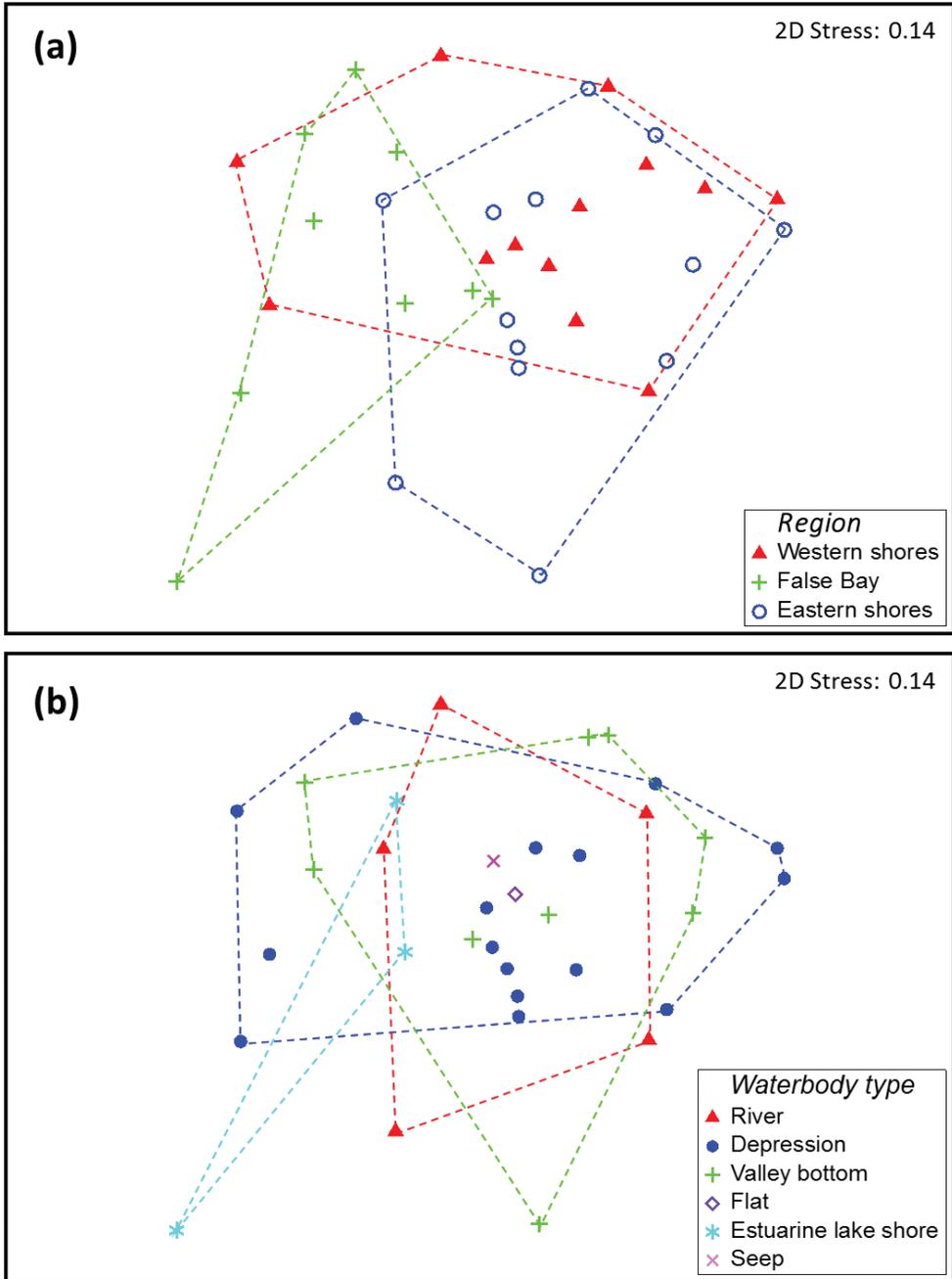


Figure 2. Multidimensional scaling (MDS) plot depicting the similarity of sites sampled in this study in terms of beetle assemblage composition. Symbols on the plot have been coded in terms of (a) region and (b) waterbody type. Convex hulls (dashed lines) have been overlaid on each plot to clarify groupings according to region/waterbody type.

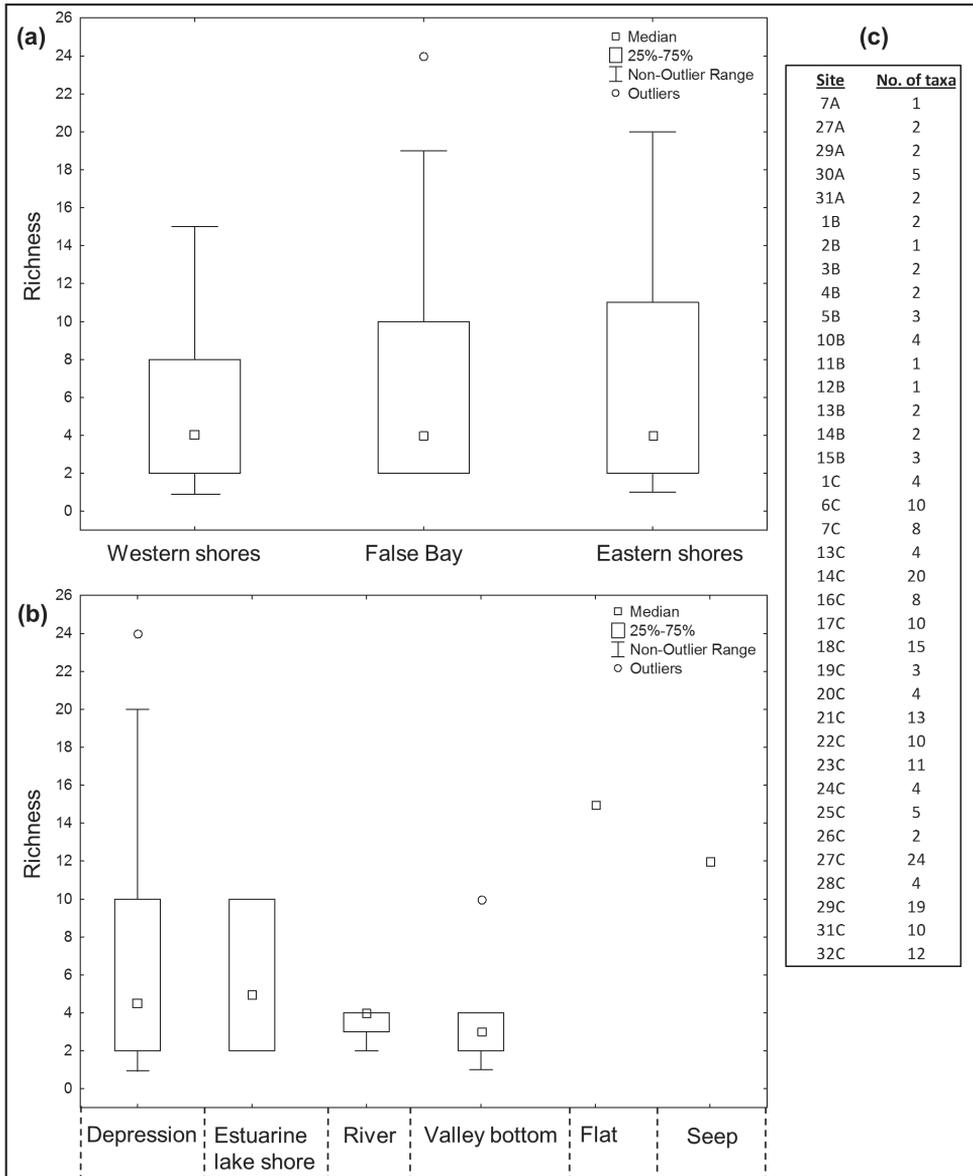


Figure 3. Box-plots comparing the median and spread of species richness (number of polyphagan taxa per site) among (a) regions and (b) waterbody types at St Lucia during the sampling period 2013–2015. The data representing number of taxa per site are also reported (c). Site numbers in (c) are coded as A (first survey–November 2013), B (second survey–July 2014) or C (third survey–January/February 2015). Kruskal-Wallis tests indicated that species richness did not vary significantly among regions ($KW-H_{2,37} = 0.9006$, $p = 0.6374$) or waterbody types ($KW-H_{5,37} = 4.2675$, $p = 0.5116$).

False Bay (site 29C with 19 species - Fig. 3c). Polyphagan species richness per site was highly correlated ($r = 0.8605$, $P < 0.001$, Fig. 4) with the richness of adepaghans sampled concurrently at the same sites (Perissinotto et al. 2016).

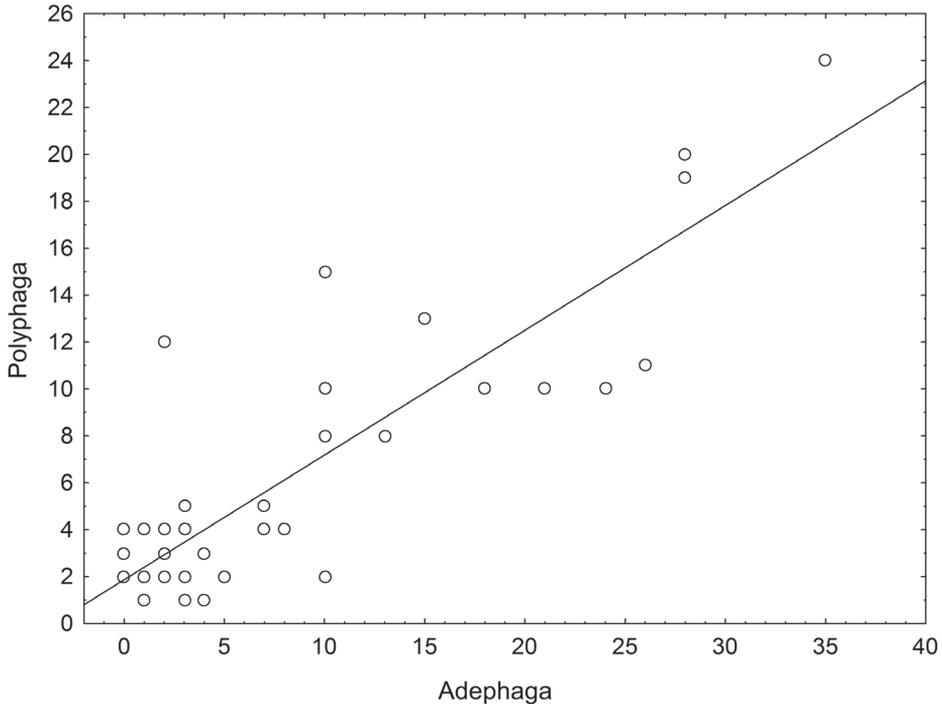


Figure 4. Scatterplot depicting the positive linear relationship ($r = 0.8605$, $P < 0.001$) between the number of taxa per site for Polyphaga (sampled in the current study) and Adephaga (concurrently sampled by Perissinotto et al. 2016).

Discussion

The dedicated surveys of the St Lucia coastal plain between 2013 and 2015 have revealed 37 aquatic polyphagan species, which predominantly reside in the small fresh-water wetlands surrounding the main lake body. Given that ca. 360 species of aquatic Polyphaga have been listed for southern Africa (Stals and de Moor 2007), the St Lucia system houses at least 10% of the aquatic polyphagan fauna of this biodiverse region. If historical records are taken into account and the seven species-level museum records (Table 3) are added to the 37 species collected during the current study, then St Lucia apparently supports at least 12% of regional diversity.

The number of Polyphaga collected at St Lucia represents approximately half the richness of hydradephagan beetles (68 taxa) reported from the same set of waterbodies by Perissinotto et al. (2016). A greater richness of the Adephaga over Polyphaga in aquatic systems has been reported elsewhere. Apenborn (2013) reported 122 species of water beetles from eight weeks of collecting effort at the Panguana research station in lowland rainforest of central Peru (Hendrich et al. 2015). Of these, around 40 belonged to the Polyphaga and 80 to the Adephaga; a ratio of ~1:2 (Polyphaga: Adephaga) as in the current study. However, this is not always the case and the ratio

appears to vary regionally. For instance, Lake Najun (approximately 100 km²) and its immediate tributaries in the Philippines produced 49 coleopteran species, of which 38 belonged to the Polyphaga, with only 10 Adephaga (Freitag and Pangan-tihon 2010). In a comprehensive checklist of aquatic beetle diversity of the Iberian Peninsula in the Mediterranean region, Ribera et al. (1998) reported 622 aquatic beetle species, however, largely due to the high regional richness of Hydraenidae (138 species), the total number of Polyphaga was 401 species, considerably outnumbering the Adephaga at 198 species. Chaco National Park and El Cachapé Wildlife Refuge in the humid Chaco Province of northern Argentina, which similarly to St Lucia is also a sub-tropical lowland plain area, albeit non-coastal, yielded 122 species, of which approximately half (60 species) belonged to the Polyphaga and the remainder were adephagans (Libonatti et al. 2013). In terms of richness, the samples were dominated by Hydrophilidae (43 species), which here even outnumbered Dytiscidae (37 species). At St Lucia Dytiscidae make up a much larger component of the fauna than Hydrophilidae, with 52 dytiscid species (Perissinotto et al. 2016) in comparison to the 27 hydrophilid species reported from the same waterbodies in the current study. This ratio is roughly in line with that reported for the Afrotropical region as a whole, given that approximately 1,060 dytiscid species have been described thus far for the region in comparison to ca. 450 hydrophilids (Jäch and Balke 2008). In their global assessment of aquatic coleopteran species diversity, Jäch and Balke (2008) report a total of approximately 7,130 polyphagan species, in comparison to 5,126 Adephaga species, a ratio of ~1.4:1 in favour of Polyphaga. In the Afrotropical region the same authors report 1,400 species of Adephaga versus 1,200 of Polyphaga (Jäch and Balke 2008), a ratio of ~1.2:1 in favour of Adephaga. One reason for the relatively low numbers of polyphagan species compared to Adephaga in the current study is the complete absence of fast running waters from this lowland region. Such habitats support large numbers of species in families such as Hydraenidae and Elmidae, including in adjacent parts of South Africa, but such beetles were entirely absent from the areas sampled in iSimangaliso Park due to the lack of suitable habitats.

The St Lucia Polyphaga were generally dominated by widespread Afrotropical taxa, with only two endemic South African species being recorded (*Paracymus amplus* Wooldridge, 1977 and *Limnebius probus* Perkins, 2015). A similar pattern (dominance of widespread taxa) is apparent at St Lucia for other invertebrate groups such as the hydradephagan beetles (Perissinotto et al. 2016), gastropods (Perissinotto et al. 2014), bivalves (Nel et al. 2012) and odonates (Hart et al. 2014). The pattern of high richness and low endemism for polyphagan water beetles at St Lucia adds further evidence to the notion that invertebrate endemism decreases, whilst diversity increases, from the south-west to the north-east of South Africa (Stals and de Moor 2007, Perissinotto et al. 2016). A new species of Hydrophilidae, *Hydrobiomorpha perissinottoi*, was discovered and described from the collections of the current study (see Bilton 2016). This species was collected in good

numbers in a variety of peripheral wetlands within the St Lucia system, although it is likely that its distribution extends throughout the broader region of Maputaland and beyond. Five species recorded in this survey are new to South Africa (Table 2), highlighting the poor state of knowledge of aquatic beetle distribution patterns in the region (see also Stals and de Moor 2007). Our survey highlights the need for further taxonomic work on some genera in the region, including *Hydrochus*, *Enochrus*, *Helochares* and *Coelostoma*, for which reliable species-level determinations are currently difficult or impossible.

In terms of the distribution of species within St Lucia, only five taxa (*Berosus viticollis* Boheman, 1851, *Enochrus (Methydrys)* sp., *Hydrochara fulvofemorata*, *Sternolophus solieri* Laporte, 1840 and *Coelostoma* sp. 2) were recorded from the margins of the lake body itself, and most taxa were instead found only in surrounding freshwater wetlands. Twelve taxa were taken with a light trap set up near the lake shore (sites 31A and 31C in November 2013 and February 2015 respectively), which captured flying adults that most likely were dispersing, perhaps from the nearby lake body. Polyphagan beetles formed a relatively distinct assemblage at False Bay, whilst the Western and Eastern Shores harbored very similar assemblages (Fig. 2a, Table 4). This same pattern was reported by Perissinotto et al. (2016) for hydradephagan beetle assemblages at St Lucia. Whilst the Eastern and Western Shores sites were generally on grassy sunlit plains, the False Bay sites occurred in dense dry woodland and were often heavily shaded, with lower resultant water temperatures (see Perissinotto et al. 2016). Although False Bay may have been distinctive in terms of its assemblage composition, the various regions of St Lucia appear to support approximately even numbers of polyphagan taxa and no region in particular was significantly elevated in terms of its species richness (Figure 3a), a finding similarly reported for the Hydradephaga of St Lucia by Perissinotto et al. (2016).

Polyphagan beetle assemblage composition did not differ between waterbody types (Fig. 2b, Table 4b), suggesting that the distribution of the species sampled, at least in the case of adults, is not affected by wetland type. Species richness also did not differ across waterbody types (Fig. 3b), further suggesting that beetles occur across the wide range of freshwater wetland type at St Lucia. That said, the two most species-rich sites sampled (20 and 24 species in sites 14C and 27C respectively) were both depression wetlands. A similar, but more pronounced pattern was reported for the Hydradephaga by Perissinotto et al. (2016), who showed that 5 of the 6 most speciose St Lucia sites were temporary depression wetlands. Polyphagan richness per site was strongly correlated with hydradephagan richness (Fig. 4) and the three most speciose sites in the current study (sites 27C, 14C and 29C) were also the most speciose for Hydradephaga. In contrast to the high beetle diversity recorded from small temporary freshwater wetlands in this study and by Perissinotto et al. (2016), other aquatic invertebrate assemblages at St Lucia have been found to be more diverse in the permanent fresh waterbodies (e.g. Hart et al. 2014) or in the estuarine lake body itself (e.g. Nel et al. 2012, Peer et al. 2014, Perissinotto et al. 2014).

Conclusions

The majority of prior aquatic research within the iSimangaliso Wetland Park has focused on the estuarine lake itself, rather than the surrounding freshwater wetlands. Our study adds evidence in addition to that of Perissinotto et al. (2016) (hydradephagan beetles) and Hart et al. (2014) (odonates) that a high biodiversity is supported by the lesser-known freshwater systems of the park, emphasizing the importance of this UNESCO World Heritage Site as a biodiversity hotspot worthy of long-term conservation efforts. Although the park itself enjoys World Heritage status, much of the St Lucia catchment has become degraded by intensive land-use practices such as commercial plantations and agriculture. These practices have resulted in a drastically reduced input of freshwater to the lake system (Perissinotto et al. 2013) that, together with a prolonged drought between 2002 and 2010, caused severe hypersalinity in the estuarine lake and a drastic loss of biodiversity (Whitfield and Taylor 2009, Carrasco and Perissinotto 2012, Bird et al. 2016). Sampling in the current study was undertaken during a relatively wet period for the system, following the almost decade-long drought and hence most of the temporary wetlands on the coastal plain were flooded. Models of future climate conditions for the region predict increased variability in rainfall and an increase in extreme climatic events (e.g. drought or flood) linked to global climate change (Shongwe et al. 2009, Davis 2011, Dallas and Rivers-Moore 2014). Further study is thus warranted into the potential effects of a changing climate and intensifying catchment land use on the invertebrate assemblages that inhabit the park's freshwater wetlands. Our results, taken together with those of Perissinotto et al. (2016), indicate that beetle biodiversity in freshwater ecosystems of the iSimangaliso Wetland Park is relatively high in a southern African context. However, to what degree this is an artefact of the generally poor state of knowledge of southern African water beetles can be better revealed by conducting comparably rigorous studies in other freshwater habitats of the region.

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Appendix I

Annotated and illustrated checklist of the Polyphaga of the Lake St Lucia system, 2013–2015.

The following list includes photographs of all species recorded during the dedicated water beetle surveys conducted by the authors during the period 2013 to 2015.

Family: Hydrochidae

Hydrochus sp. 1

Remarks. The Afrotropical species of this genus are in need of revision.

Distribution. Range unknown. Afrotropical.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in July 2014 and January/February 2015. Previously recorded in fresh water wetlands by Vrdoljak (2004) in 2002/2003.



Figure 5. *Hydrochus* sp. 1
2.5 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Hydrochus sp. 2

Remarks. The Afrotropical species of this genus are in need of revision.

Distribution. Range unknown. Afrotropical.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015. Previously recorded in fresh water wetlands by Vrdoljak (2004) in 2002/2003.



Figure 6. *Hydrochus* sp. 2
2.6 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Hydrochus sp. 3

Remarks. The Afrotropical species of this genus are in need of revision.

Distribution. Range unknown. Afrotropical.

St Lucia records. Recorded at Western Shores in January/February 2015.



Figure 7. *Hydrochus* sp. 3
2.4 mm, iSimangaliso Wetland Park, Western
Shores (site 18), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Previously recorded in fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.

Family: Spercheidae

***Spercheus cerisyi* Guérin-Méneville, 1842**

Synonyms. *Spercheus crenaticollis* Régimbart, 1906, *Sphercheus* [!] *capicola* Péringuey, 1829, *Spercheus cerisyi* var. *diminutus* Hebauer, 1997

Remarks. Ponds and other lentic waters, in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa and Madagascar; reaching the Palearctic in Egypt, Iraq and Israel.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015. Previously recorded at Dukandlovu by SANC – years not specified.



Figure 8. *Spercheus cerisyi* Guérin-Méneville, 1842
2.4 mm, iSimangaliso Wetland Park, Eastern Shores (site 14), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Spercheus senegalensis* Castelnau, 1832**

Synonyms. *Spercheus sulcatus* Gory, 1834, *Sphercheus* [!] *algoensis* Péringuey, 1892, *Spercheus distinguendus* Fairmaire, 1893.

Remarks. Ponds and other lentic waterbodies rich in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa and Madagascar; apparently reaching the Palearctic in Turkey.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015. Previously recorded at St Lucia and Dukandlovu by SANC – years not specified.



Figure 9. *Spercheus senegalensis* Castelnau, 1832
4.5 mm, iSimangaliso Wetland Park, False Bay (site 29), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Family: Hydrophilidae***Amphiops globus* Erichson, 1843**

Remarks. Ponds and other lentic waterbodies.

Distribution. Widespread to Western, Central and Eastern Africa. Also reported from China.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.

***Amphiops senegalensis* (Laporte, 1840)**

Synonyms. *Amphiops lucidus* Erichson, 1843, *Cyprimorphus compressus* Fairmaire, 1873, *Amphiops Abeillei* Guillebeau, 1896, *Amphiops lucidus* var. *abeillei* Guillebeau, 1896, *Amphiops lasioides* Régimbart, 1903.

Remarks. Ponds and other lentic waterbodies.

Distribution. Widespread to Western, Central, Northern and Eastern Africa; reaching the Palearctic in Egypt and Morocco.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in July 2014 and January/February 2015.

***Amphiops ubligi* Hebauer, 1995**

Remarks. Found in dense vegetation in a small wetland at St Lucia.

Distribution. Namibia, Botswana and Zambia. New record for South Africa.

St Lucia records. Recorded at Eastern Shores in January/February 2015.



Figure 10. *Amphiops globus* Erichson, 1843
5.1 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.



Figure 11. *Amphiops senegalensis* Laporte, 1840
5.0 mm, iSimangaliso Wetland Park, Eastern
Shores (site 14), February 2015
DT Bilton, MS Bird & R Perissinotto leg.



Figure 12. *Amphiops ubligi* Hebauer, 1995
3.5 mm, iSimangaliso Wetland Park, Eastern
Shores (site 14), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Allocotocerus* sp.**

Remarks. Species-level identification requires comparison with types.

Distribution. Range unknown.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013, July 2014 and January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 13. *Allocotocerus* sp.

4.3 mm, iSimangaliso Wetland Park, False Bay (site 27), February 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Berosus cuspidatus* Erichson, 1843**

Synonyms. *Berosus bispinosus* Boheman, 1851, *Berosus acutispina* Fairmaire, 1869, *Berosus cuspidatus* ssp. *acutispina* Fairmaire, 1869, *Berosus gracilispina* Régimbart, 1906.

Remarks. Ponds and lagoons, particularly with exposed substrate and some mineralisation/salinity.

Distribution. Widespread to Western, Central and Eastern Africa and Madagascar and the Seychelles; reaching the Palearctic in Egypt.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015. Previously recorded at False Bay by Day et al. (1954) and Millard and Broekhuysen (1970) in 1948.



Figure 14. *Berosus cuspidatus* Erichson, 1843

5.2 mm, iSimangaliso Wetland Park, False Bay (site 29), February 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Berosus viticollis* Boheman, 1851**

Remarks. Lentic waters, particularly with exposed substrates.

Distribution. Widespread to Western, Central and Eastern Africa and Madagascar.

St Lucia records. Recorded at Western Shores and False Bay in November 2013.



Figure 15. *Berosus viticollis* Boheman, 1851

2.8 mm, iSimangaliso Wetland Park, False Bay (site 29), February 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Regimbartia nilotica* (Sharp, 1903)**

Synonyms. *Volvulus compressus* Régimbart, 1906, *Regimbartia compressa* (Régimbart, 1906).

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa; reaching the Palearctic in Egypt.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013 and January/February 2015.



Figure 16. *Regimbartia nilotica* (Sharp, 1903)
5.4 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Regimbartia obsoleta* (Régimbart, 1906)**

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.



Figure 17. *Regimbartia obsoleta* (Régimbart, 1906)
3.8 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Laccobius uhligi* Gentili, 1995**

Remarks. Seepages over peat beside lagoon at St Lucia.

Distribution. Namibia (Caprivi Strip) and Botswana (Okavango). New record for South Africa.

St Lucia records. Recorded at Eastern Shores in January/February 2015.



Figure 18. *Laccobius uhligi* Gentili, 1995
1.8 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015

DT Bilton, MS Bird & R Perissinotto leg.

***Paracymus amplus* Wooldridge, 1977**

Remarks. Lentic waters, in vegetation.

Distribution. A species currently only known from South Africa.

St Lucia records. Recorded at Eastern Shores in January/February 2015.



Figure 19. *Paracymus amplus* Wooldridge, 1977
2.5 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Paracymus exiguus* Wooldridge, 1977**

Remarks. Lentic waters, in vegetation.

Distribution. Described from Zimbabwe. New record for South Africa.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.

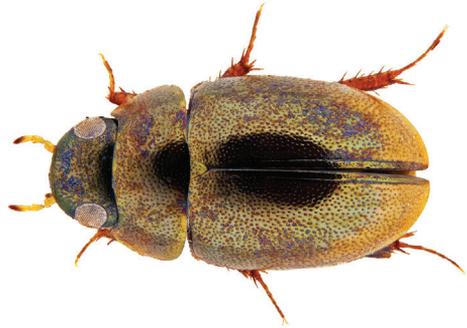


Figure 20. *Paracymus exiguus* Wooldridge, 1977
2.5 mm, iSimangaliso Wetland Park, False Bay
(site 29), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Paracymus pisanus* Balfour-Browne, 1954**

Remarks. Lentic waters, in vegetation.

Distribution. South Africa, Namibia and Botswana.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.



Figure 21. *Paracymus pisanus* Balfour-Browne, 1954
1.9 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Hydrobiomorpha perissinottoi* Bilton, 2016**

Remarks. A new species, first detected during this survey. Most close morphologically to *H. occidentalis* Balfour-Browne, 1939 from Nigeria and Sudan.

Distribution. Currently only recorded from St Lucia – wider distribution unknown.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.



Figure 22. *Hydrobiomorpha perissinottoi* Bilton, 2016

18.5 mm, iSimangaliso Wetland Park, Eastern Shores (site 22), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Hydrochara elliptica* (Fabricius, 1801)**

Synonyms. *Hydrochaeres ellipticus* (Fabricius, 1801), *Hydrous uniformis* Fairmaire, 1869, *Hydrophilus fulvo-femorata* var. *uniformis* (Fairmaire, 1869).

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to western, central and eastern Africa and Madagascar.

St Lucia records. Recorded at False Bay in November 2013. Previously recorded at Dukandlovu Pan by the authors and deposited at UKZN in 2012.

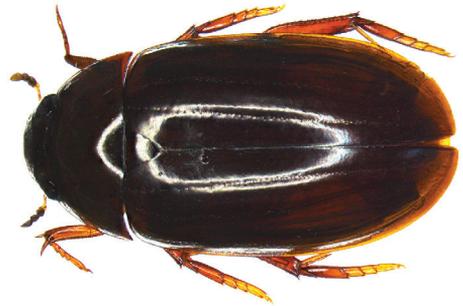


Figure 23. *Hydrochara elliptica* (Fabricius, 1801)

16.0 mm, iSimangaliso Wetland Park, False Bay (site 31), November 2013
MS Bird & R Perissinotto leg.

***Hydrochara fulvofemorata* (Fairmaire, 1869)**

Remarks. Lentic waters, in vegetation.

Distribution. Mozambique to Eastern Africa and Madagascar. New record for South Africa, first reported by Bilton (2016).

St Lucia records. Recorded at Western Shores and False Bay in November 2013 and January/February 2015.



Figure 24. *Hydrochara fulvofemorata* (Fairmaire, 1869)

16.2 mm, iSimangaliso Wetland Park, False Bay (site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Hydrophilus aculeatus* (Solier, 1834)**

Synonyms. *Hydrophilus spinipennis* Gory, 1834, *Hydrophilus armatus* Castelnau, 1840, *Hydrophilus lugubris* Motschulsky, 1845, *Hydrophilus aegyptiacus* Peyron, 1856.

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa, the Mascarenes and Arabia; reaching the Palaearctic in Egypt, Iran, Israel, Syria and Turkey.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay November 2013, July 2014 and January/February 2015. Previously recorded at False Bay by the authors and deposited at UNKZ in 2012.

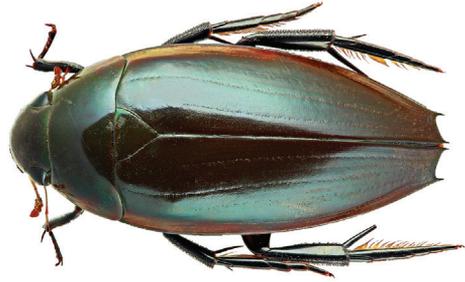


Figure 25. *Hydrophilus aculeatus* (Solier, 1834)
37.0 mm, iSimangaliso Wetland Park, Eastern Shores (site 14), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Sternolophus solieri* Laporte, 1840**

Synonyms. *Sternolophus rufipes* Solier, 1834, *Helobius noticolis* Mulsant, 1851, *Hydrous aeratus* Reiche & Saulcy, 1854, *Hydrous graecus* Baudi, 1864, *Sternolophus punctatulus* Schaufuss, 1883.

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to Western, Central and Eastern Africa, Madagascar, the Cape Verdes, the Comoros; reaching the Palaearctic in Algeria, Egypt, Greece and Israel.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013 and January/February 2015. Previously recorded at False Bay by the authors and deposited at UKZN in 2008.



Figure 26. *Sternolophus solieri* Laporte, 1840
9.9 mm, iSimangaliso Wetland Park, False Bay (site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

Enochrus (Methydrus) sp.

Remarks. Lentic waters, in vegetation. African fauna requires revision.

Distribution. Unknown.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013, July 2014 and January/February 2015.



Figure 27. *Enochrus (Methydrus) sp.*
2.8 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Chasmogenus cf. patrizii* (Balfour-Browne, 1948)**

Remarks. Lentic waters, in vegetation. African fauna requires revision before certain identification can be reached.

Distribution. Widespread to Central and Eastern Africa.

St Lucia records. Recorded at Eastern Shores and False Bay in January/February 2015.



Figure 28. *Chasmogenus cf. patrizii* (Balfour-Browne, 1948)
3.4 mm, iSimangaliso Wetland Park, Eastern
Shores (site 23), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Helochares dilutus* (Erichson, 1843)**

Synonyms. *Helochares niloticus* Sharp, 1903.

Remarks. Lentic waters, in vegetation. Records from South Africa, Namibia, Madagascar and the Mascarenes have been referred to ssp. *consputus* Boheman, 1851.

Distribution. Widespread to Western, Central and Eastern Africa and Madagascar.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in July 2014 and January/February 2015. Previously recorded at St Lucia and Dukandlovu by SANC – years not specified.



Figure 29. *Helochares dilutus* (Erichson, 1843)
6.8 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Helochares longipalpis* (Murray, 1859)**

Remarks. Lentic waters, in vegetation.

Distribution. Widespread to Western, central and Eastern Africa; reaching the Palaearctic in Egypt and Israel.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in July 2014 and January/February 2015. Previously recorded at St Lucia and Dukandlovu by SANC – years not specified.



Figure 30. *Helochares longipalpis* (Murray, 1859)
7.0 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Helochares* sp. 1**

Remarks. Lentic waters, in vegetation. The African *Helochares* fauna requires a thorough revision.

Distribution. Unknown.

St Lucia records. Recorded at Eastern Shores and False Bay in July 2014 and January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 31. *Helochares* sp. 1
4.8 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Helochares* sp. 2**

Remarks. Lentic waters, in vegetation. The African *Helochares* fauna requires a thorough revision.

Distribution. Unknown.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013, July 2014 and January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 32. *Helochares* sp. 2
2.9 mm, iSimangaliso Wetland Park, False Bay
(site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Coelostoma* sp. 1**

Remarks. The African fauna of this genus requires revision.

Distribution. Unknown.

St Lucia records. Recorded at Eastern Shores and False Bay in January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 33. *Coelostoma* sp. 1
4.5 mm, iSimangaliso Wetland Park, Catalina Bay
(site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Coelostoma* sp. 2**

Remarks. The African fauna of this genus requires revision.

Distribution. Unknown.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in November 2013, July 2014 and January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 34. *Coelostoma* sp. 2
6.3 mm, iSimangaliso Wetland Park, Eastern
Shores (site 23), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Coelostoma* sp. 3**

Remarks. The African fauna of this genus requires revision.

Distribution. Unknown.

St Lucia records. Recorded at Eastern Shores in January/February 2015. Previously recorded at fresh water wetlands on the Eastern Shores of Lake St Lucia by Vrdoljak (2004) in 2002/2003.



Figure 35. *Coelostoma* sp. 3
4.8 mm, iSimangaliso Wetland Park, Eastern
Shores (site 23), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Cercyon dieganus* Régimbart, 1903**

Remarks. In wet decaying vegetable debris in the margins of lentic waterbodies.

Distribution. Widespread in Afro-tropical Region, including Madagascar.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.



Figure 36. *Cercyon dieganus* Régimbart, 1903
2.6 mm, iSimangaliso Wetland Park, Eastern Shores (site 23), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Family: Hydraenidae***Hydraena cooperi* Balfour-Browne, 1954**

Remarks. Shallow margins of lentic and lotic waters.

Distribution. Widespread in South Africa and also recorded from Angola and Namibia.

St Lucia records. Recorded at Western Shores, Eastern Shores and False Bay in January/February 2015.



Figure 37. *Hydraena cooperi* Balfour-Browne, 1954
1.5 mm, iSimangaliso Wetland Park, Catalina Bay (site 32), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Limnebius probus* Perkins, 2015**

Remarks. Pond margins.

Distribution. Eastern South Africa. Species recently described, but so far only known from South Africa.

St Lucia records. Recorded at False Bay in January/February 2015.



Figure 38. *Limnebius probus* Perkins, 2015
0.9 mm, iSimangaliso Wetland Park, False Bay (site 27), January 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Aulachochthebius cf. continentalis* (Orchymont, 1929)**

Remarks. Pond margins. Afrotropical species of the genus currently in revision (Perkins, pers. comm.).

Distribution. Kenya. Most likely new to South Africa, regardless of species.

St Lucia records. Recorded at Eastern Shores and False Bay in January/February 2015.



Figure 39. *Aulachochthebius cf. continentalis* (Orchymont, 1929)

1.2 mm, iSimangaliso Wetland Park, Eastern Shores (site 21), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

***Ochthebius andronicus* Orchymont, 1948**

Remarks. Pond margins.

Distribution. Widespread in Southern and Eastern Africa.

St Lucia records. Recorded at Eastern Shores and False Bay in January/February 2015.



Figure 40. *Ochthebius andronicus* Orchymont, 1948

2.0 mm, iSimangaliso Wetland Park, Eastern Shores (site 21), February 2015
DT Bilton, MS Bird & R Perissinotto leg.

Family: Curculionidae

***Pseudobagous cf. longulus* (Gyllenhal, 1836)**

Remarks. In vegetation in lentic waterbodies.

Distribution. Widespread in Southern Africa.

St Lucia records. Recorded at Eastern Shores and False Bay in July 2014 and January/February 2015.



Figure 41. *Pseudobagous cf. longulus* (Gyllenhal, 1836)

5.0 mm, iSimangaliso Wetland Park, Eastern Shores (site 13), July 2014
MS Bird leg.

The genus *Alaolacon* Candèze, a senior synonym of the genus *Eumoeus* Candèze (Coleoptera, Elateridae, Agrypninae)

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Abstract

Alaolacon Candèze, 1865 is found to be a senior synonym of *Eumoeus* Candèze, 1874, *Luzonicus* Fleutiaux, 1916 and *Tharopsides* Fleutiaux, 1918. *Alaolacon* is represented by *A. bakeri* (Fleutiaux, 1916), **comb. n.**, *A. candezei* Fleutiaux, 1928, *A. cyanipennis* Candèze, 1865, *A. fujiokai* **sp. n.**, *A. griseus* Candèze, 1874, *A. megalopus* **sp. n.**, *A. murrayi* (Candèze, 1874), **comb. n.**, and *A. philippinensis* **nom. n.** This genus is redescribed based on the descriptions of three species, *A. candezei*, *A. fujiokai*, and *A. megalopus* as well as the examination of the holotypes of *A. cyanipennis* and *A. murrayi* **comb. n.** Males of the genus *Alaolacon* exhibit 12-segmented and bilabellate antennae, and the females exhibit 11-segmented and subpectinate antennae. A key to species is provided.

Keywords

Eumaeus, Hemirhipini, *Luzonicus*, new species, Oriental region, replacement name, taxonomy, *Tharopsides*

Introduction

Candèze (1865) established the monotypic genus *Alaolacon* for *A. cyanipennis* from the Peninsular Malaysia. *Alaolacon griseus* Candèze, 1874 from Thailand and *A. candezei* Fleutiaux, 1928 from Banggi Island, Malaysia (near Borneo) were described later. All specimens described in this genus were females, with 11-segmented and subpectinate antennae, and the males were undescribed. Candèze (1874) established genus *Eumoeus* for one species, *E. murrayi*, from India from a male with 12-segmented and biflabellate antennae. He argued that *Eumoeus* was similar to *Alaolacon*, although they had extremely different antennae, and suggested that *Alaolacon* should be combined with *Eumoeus* if its male had biflabellate antennae.

Fleutiaux (1916) established *Luzonicus*, containing only *L. bakeri* from the Philippines, from a female specimen with 11-segmented and moniliform antennae. Fleutiaux (1918) later established *Tharopsides* including two species, *T. harmandi* and *T. bakeri* from Thailand and the Philippines respectively, from males possessing 12-segmented and biflabellate antennae. Fleutiaux, (1940) stated that *Eumoeus* was a junior homonym of *Eumaeus* Hübner, 1816 of Lepidoptera, and used *Tharopsides* as the replacement name. Fleutiaux (1947) subsequently stated that *Tharopsides* was a junior synonym for *Luzonicus*, and that antennal differences were sexual dimorphism. However, Casari-Chen (1993, 1994) and Casari (2008) treated *Eumoeus* as a valid name and a monotypic genus, making no mention about the treatments of Fleutiaux (1940, 1947). This paper reviews the taxonomy of these four genera of Hemirhipini and of five of eight included species in order to resolve this confusion.

Materials

Depositories of the type specimens and non-type specimens examined are as follows:

- BMNH** The Natural History Museum, London,
- MNHN** Muséum national d'Histoire naturelle, Paris (Edmond Fleutiaux collection),
- IRSNB** Institut Royal des Sciences Naturelles de Belgique, Brussels,
- ELKU** Entomological Laboratory, Kyushu University, Fukuoka.

A generic description of *Alaolacon* was made from the study of the type specimens of *A. cyanipennis* Candèze, 1865, *A. candezei* Fleutiaux, 1928, *A. murrayi* (Candèze, 1874) comb. n. (= *Eumoeus murrayi*) and two new species described here. Species descriptions of *A. cyanipennis* and *A. murrayi* are not provided as they are adequately described in Casari-Chen (1993).

We could not find the types of two species, *A. bakeri* (Fleutiaux, 1916) comb. n. (= *Luzonicus bakeri*) and *A. philippinensis* comb. n. (= *Tharopsides bakeri* Fleutiaux, 1918) in the collections of BMNH, IRSNB nor the MNHN and have not examined these species. It was not possible to prepare a description of *A. griseus* Candèze, 1874.

Methods

Photographs of specimens were taken by a single-lens reflex camera (Canon EOS 70D) with a macro lens (Canon macro photo lens MP-E 65mm), and then images taken in a series of focal planes were combined using CombineZM 1.0.0 software (Alan Hadley, United Kingdom). Micrographs were prepared using a scanning electron microscope (SEM: Hitachi S-3000N) without gold coating.

Most structures were observed under a stereo microscope (Olympus-SZX9). Measurements are in millimeters and were made with a micro ruler (MR-2, Kenis Limited, Ôsaka; minimum scale value: 0.05 mm). Specimens were softened in warm water. The pregenital segments and genitalia extracted from the abdomen were soaked in 10% KOH solution (room temperature, male: 2 hours, female: 48 hours). The parts were cleaned in 30% ethanol (10 min) and dehydrated in 99.5% ethanol (5 min) and then mounted in glycerin on microscope slides, except the female genitalia, which were examined in water and then mounted in glycerin. A transmission microscope (Nikon Y-IDT) with a *camera lucida* was used to examine slides and for drawing. Morphological terminology follows Calder (1996), and Casari-Chen (1993) and Costa et al. (2010) in part. Photographs and drawings were edited with image editing software (Adobe Photoshop 7.0).

The following abbreviations are used:

BL	body length from head to elytral apices
BW	the maximum body width
MIE	the minimum distance between the eyes
MAE	the maximum distance across the eyes
OI	Ocular index: $MIE/MAE \times 100$
PL	the maximum pronotum length including posterior angles
PML	length of the midline of pronotum
PW	the maximum pronotum width including posterior angles
PI	Pronotam index: $PL/PW \times 100$
EL	the maximum elytra length
EW	the maximum elytra width
EI	Elytra index: $EL/EW \times 100$

Taxonomy

Genus *Alaolacon* Candèze, 1865

Alaolacon Candèze, 1865: 13 (original description; type species: *Alaolacon cyanipennis* Candèze, 1865; by monotypy; in Mélanactides); Gemminger and Harold 1869: 1498 (catalogue of Coleoptera); Candèze 1874: 114 (in tribe Alaites); Candèze 1891: 29 (short description; in tribe Alaites), 241 (index); Schwarz 1906: 316 (cat-

alogue); Hyslop 1921: 625 (type species of genera of Elateridae); Schenkling 1925: 40 (catalogue); Fleutiaux 1926: 102 (catalogue); Fleutiaux 1928: 177 (description); Casari-Chen 1993: 227 (description; removed from Hemirhipini); Casari 2008: 166 (key to genera of Hemirhipini; replaced in Hemirhipini).

Eumoeus Candèze, 1874: 113 (original description; type species: *Eumoeus murrayi* Candèze, 1874; by monotypy; in tribe Alaites), 214 (as “*Eumaeus*”; index); Candèze 1891: 29 (short description; in tribe Alaites), 243 (index); Schwarz 1906: 32 (key to genera of Hemirhipini), 40 (catalogue); Hyslop 1921: 645 (type species); Schenkling 1925: 51 (as “*Eumaeus*”; catalogue); Fleutiaux 1928: 178 (as “*Eumaeus*”; comments); Fleutiaux 1947: 306 (as junior homonym of *Eumaeus* Hübner, 1816 (Lepidoptera)); Casari-Chen 1993: 241 (description; removed from Hemirhipini); Casari 2008: 164 (key to genera of Hemirhipini; replaced in Hemirhipini) **Syn. n.**

Luzonicus Fleutiaux, 1916: 232 (original description; type species: *Luzonicus bakeri* Fleutiaux, 1916; by monotypy; in Corymbitinae); Schenkling 1927: 405 (catalogue); Fleutiaux 1947: 306 (key to genera of Oxynopterinae; description); Tarnawski 2001: 306 (catalogue of Ctenicerini, Athoinae).

Tharopsides Fleutiaux, 1918: 235 (original description; type species: *Tharopsides harmandi* Fleutiaux, 1918); Hyslop 1921: 671 (type species); Fleutiaux 1924: 176 (reprinting of original description); Fleutiaux 1928: 178 (taxonomic comments); Schenkling 1927: 509 (catalogue); Fleutiaux 1940: 40 (as replacement name for *Eumoeus* Candèze, 1874; in Hemirhipinae); Fleutiaux 1947: 306 (as synonym of *Luzonicus* Fleutiaux, 1916).

Diagnosis. Setae flat, wider at midlength than base, with longitudinal micro carinae (Figs 36, 37); interspaces between punctures greater than puncture diameter except for narrower interspaces on head and pronotum; supra-antennal carinae not continuous across frons; frontoclypeal region gradually sloping to base of labrum; antennae 12-segmented and biflabellate in male (Figs 23, 42) or 11-segmented and subpectinate in females (Fig. 5); mandibles bidentate; hypomeron with mesal edge with impunctate ridge next to prosternal suture and carinate anterolaterad (Figs 8, 26, 45: arrow), posterior edge with two angles near mid-length (Figs 9, 27, 46: arrows); scutellum widest posteriorly or with parallel sides; elytral intervals convex; hind wings with vein r₄ translucent (Figs 11, 29, 48); parameres of male aedeagus not fused and without acute lateral subapical barb (Figs 34, 35, 53, 54).

Redescription. Adult. *Body* (Figs 1, 17, 20, 38, 55) 11–24 mm; surface smooth, with or without metallic luster on elytra; interspaces between punctures greater than puncture diameter except for narrower interspaces on head and pronotum. Vestiture. Setae flat, wider at midlength than base, with longitudinal micro carinae (Fig. 36); carinae converge at apex, apices acute or transverse (Fig. 37). **Male.** Antennomeres III–XII with setae filiform ventrally.

Head (Figs 4, 22, 41) depressed longitudinal medially, depression becoming narrow and shallow posteriorly. Frontal depression moderate (Figs 4, 41) to deep

(Figs 22, 56). Eyes small to very large (OI: 44–74). Supra-antennal carina not continuous across frons. Frontoclypeal region gradually sloping to base of labrum. Labrum subtrapezoidal; anterior angles rounded. Antennae not reaching pronotum posterior lateral apices; antennomere I cylindrical; antennomere II shortest. **Male** (Figs 23, 42). 12-segmented; antennomeres III–XI biflabellate; antennomere XII blade-like. **Female** (Fig. 5). 11-segmented; antennomere III subpectinate to trapezoidal, longer than wide ($1.2\text{--}1.4 \times$ as long as wide); antennomeres IV–X pectinate, shorter than wide (less than $0.6 \times$ as long as wide); antennomere XI elliptical. Mandibles bidentate (Fig. 6). Labium (Figs 6, 24, 43); mentum membranous in anterior part; prementum widest anteriorly, with anterior margin fringed with short setae. Apical maxillary palpomere $1.3\text{--}1.8 \times$ as long as wide.

Prothorax shorter to longer than wide, widest posteriorly or at mid-length. Pronotum with anterior angle bisinuate (Figs 1, 17, 20, 56) or rounded (Fig. 38); hind angles unicarinate; median longitudinal depression present extending at almost all pronotal length (Figs 4, 22, 56) or at pronotal anterior half (Fig. 41). Hypomeron concave; impunctate posterad; anterior angles rounded (Fig. 8) to acute (Figs 26, 45); external margins of depressions for reception of forelegs not carinate; mesal edge with elevated impunctate ridge next to prosternal suture, carinate anterolaterad (Figs 8, 26, 45: arrow); posterior edge with two angles near midlength (Figs 9, 27, 46: arrows). Prosternum produced forwards, exceeding anterior angles of pronotum; prosternal spine inclined dorsally behind procoxae weakly (at less than 10 degrees, Fig. 7) to strongly (more than 10 degrees, Figs 25, 44).

Mesothorax. Scutellum longer than wide; anterior margin straight, well defined by wrinkled band; sides concave or straight, widest posteriorly (Figs 10, 28, 56) or parallel (Fig. 47); rounded posterad. Mesosternum and metasternum not fused. Mesosternal cavity with median shiny band formed by dense yellow setae (Fig. 2, 21, 39). Mesepisternum centrally impunctate. Mesepisternum and mesepimeron reaching mesocoxal cavity. Metasternum with shallow median longitudinal groove. Elytra with striae impressed and with punctures; apex rounded. Hind wings with vein r_4 translucent; bear or lack wedge cell; cross vein between veins MP4 and CuA2, located at contact point between veins MP3 and MP4 (Figs 11, 18), or anterad to the contact point (Figs 29, 48). Legs with simple tarsomeres and tarsal claws. Tibial spurs present. Tarsomeres II–IV short, tarsomere V longest.

Abdomen. **Male.** Tergite VIII shorter than wide (Fig. 30) or longer than wide (Fig. 49). Sternite VIII (Figs 31, 50) wide-rectangular. Tergite IX (Figs 32, 51) wide; posterior margin notched medially. Tergite \times (Figs 32, 51) semicircular. Sternite \times attached to sternite IX (Figs 33, 52). **Female.** Tergite VIII (Fig. 12) truncate apically. Sternite VIII (Fig. 13) with spiculum ventrale robust, with apex concave or rounded.

Genitalia. **Male.** Aedeagus (Figs 34, 35, 53, 54, 57) with parameres unfused, without acute lateral subapical barb, with apical parts expanded elliptically. **Female.** Ovipositor (Fig. 14) stout. Coxites (Figs 15) without styli. Vagina and bursa copulatrix without sclerotized structures (Figs 16).

Larvae and pupae. Unknown.

Distribution. Oriental Region: India, Thailand, Vietnam, Indonesia (Sumatra, Java), Malaysia (Peninsular Malaysia, Borneo), the Philippines (Mindanao Is., Luzon Is.).

Bionomics. Nothing is known of the life history and ecology.

Alaolacon candezei Fleutiaux, 1928

Figures 1–16

Alaolacon candezei Fleutiaux, 1928: 177 (original description; type locality: Malaysia, East Malaysia (Sabah), Banggi Island).

Type material. Holotype. Female, Banggi Island (located off the northern coast of Borneo), Sabah, Malaysia, Waterstradt leg. [MNHN] (Fig. 3). Label data: “TYPE”; [female symbol]; “Banguey/ Borneo/ Waterstradt” “= cyanipennis Cand.?!/ Collection FLEUTIAUX”; “Alaolacon/ candezei/ Fleut. type/ Collection FLEUTIAUX”; “Alaolacon/ candezei Fleut./ COLLECTION FLEUTIAUX”; “Muséum paris/ Coll./ E. Fleutiaux”.

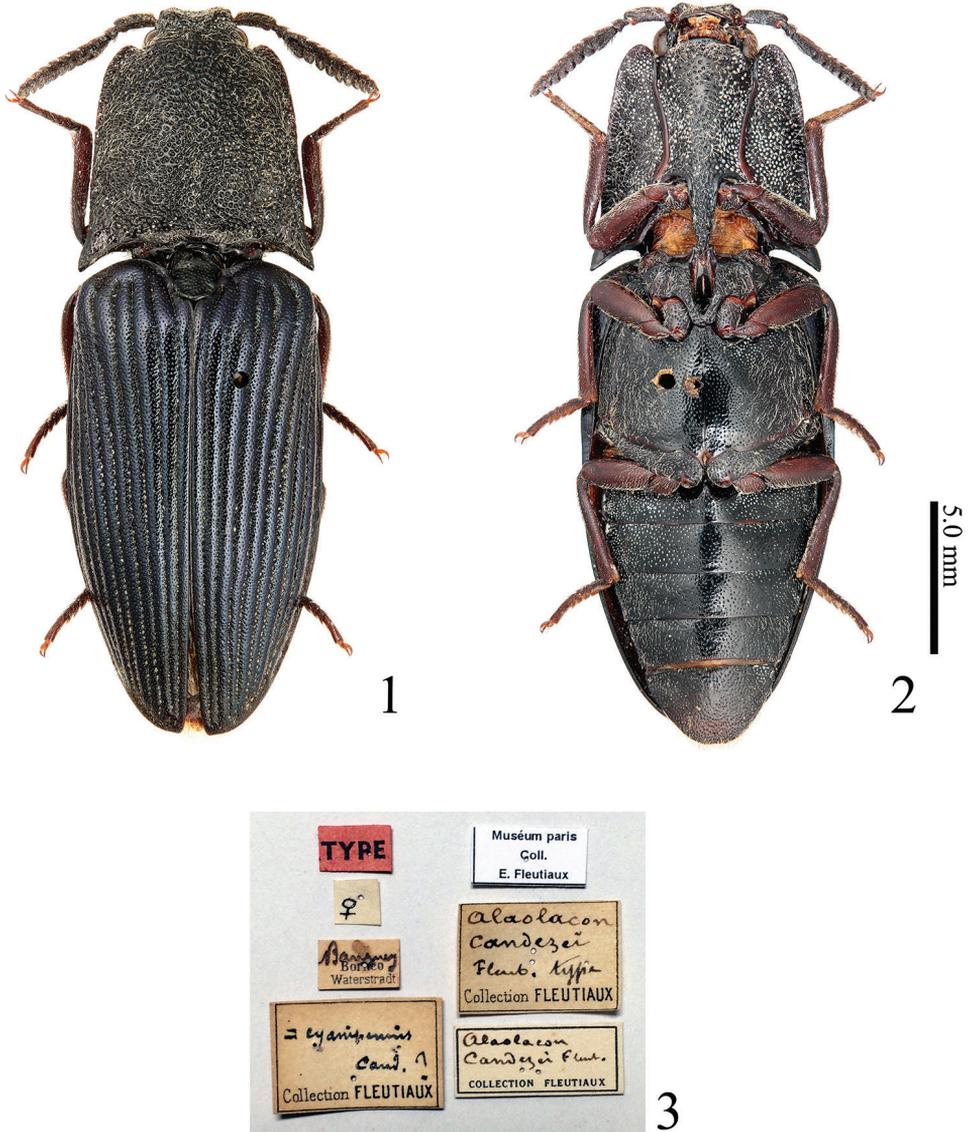
Diagnosis. Body black, elytra blue and with metallic luster, legs red-black; setae white; frontal depression moderate; eye small; female antennomere III subpectinate, $1.2 \times$ as long as wide; prothorax almost as long as wide, widest posteriorly; pronotum with anterior angles bisinuate, median longitudinal depression shallow, not reaching anterior margin or base, punctate; anterior angles of hypomeron rounded; prosternal spine inclined weakly behind procoxae; scutellum concave laterally, widest near posterior $2/5$; hind wings without wedge cell, with cross vein between veins MP4 and CuA2 located at contact point between veins MP3 and MP4; female sternite VIII with apex concave.

Measurements. BL: 24.0, BW: 8.35, MIE: 2.56, MAE: 3.47, OI: 74, PL: 7.64, PML: 6.67; PW: 7.70, PI: 99, EL: 15.7, EW: 8.35, EI: 188.

Redescription of female. *Body* (Figs 1, 2) shiny; elytra with weak metallic luster. *Color.* Body black; mouth-parts brown, mandible black, galea and lacinia orange; elytra black-blue; pronotosternal sutures and legs red-black; tarsal claws yellow-brown. *Hairs.* Body covered with white flatted setae; antennomere I and legs with intermixed brown and white setae; antennomeres II–XI with brown setae. (Most setae of elytra lost.)

Head. Frontal depression moderate (Fig. 4). Eyes small. Antennomere II conical; antennomere III longest, subpectinate, $1.2 \times$ as long as wide, $3.0 \times$ times as long as II; apical half part of antennomere XI thinner than its basal half part (Fig. 5: dotted line). Apical maxillary palpomere $1.6 \times$ as long as wide (Fig. 6).

Prothorax almost as long as wide, widest posteriorly; hind angles straight posteriorly. Pronotum with anterior angle bisinuate; median longitudinal depression shallow, not reaching anterior margin or base, punctate. Hypomeron with anterior angles rounded (Fig. 8). Prosternal spine inclined weakly (at 8 degrees) behind procoxae (Fig. 7). Scutellum (Fig. 10) $1.2 \times$ as long as wide, concave laterally, widest near posterior $2/5$. Hind wings with cross vein between veins MP4 and CuA2 apparent, not completely connected with CuA2, located at contact point between veins MP3 and

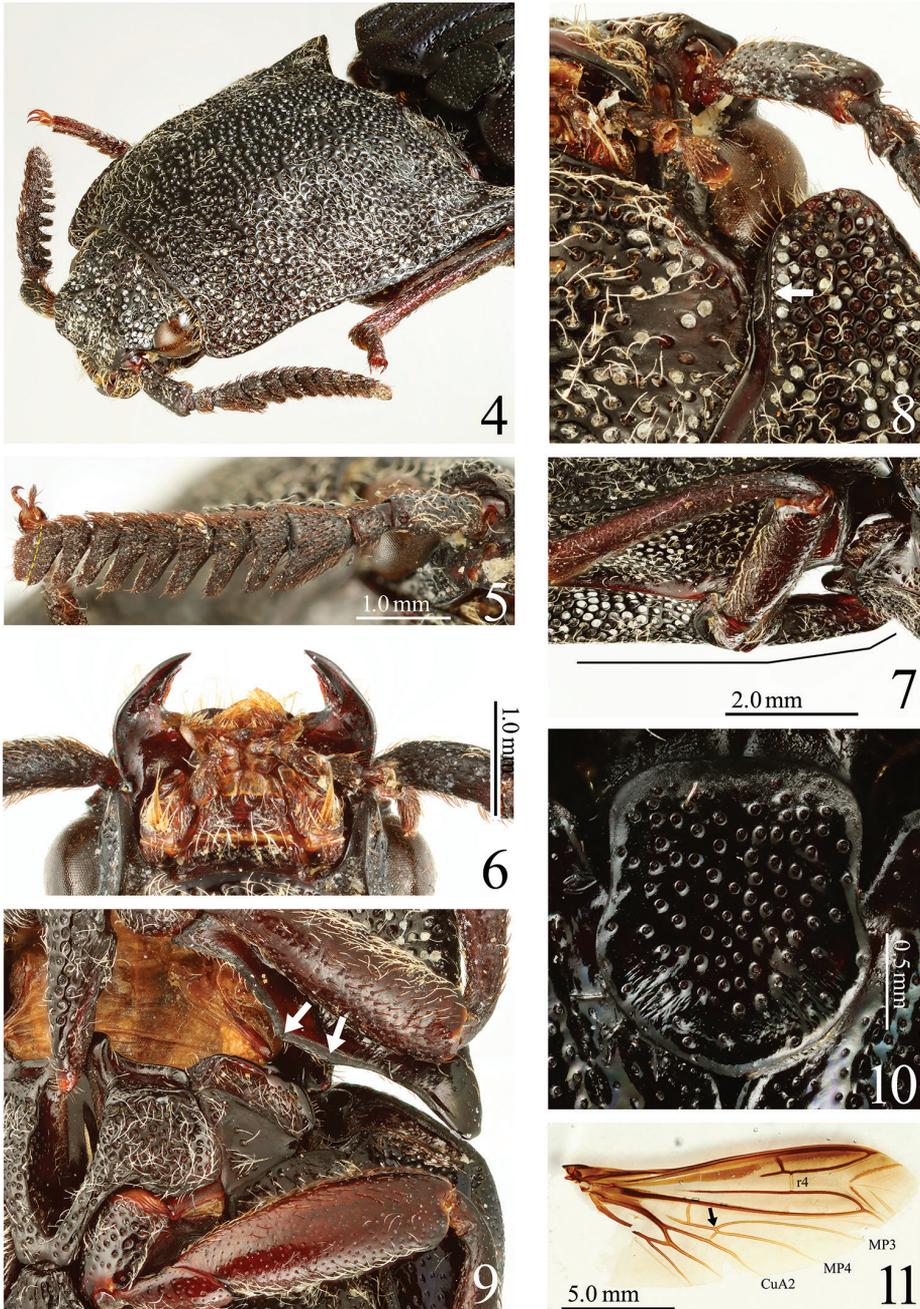


Figures 1–3. *Alaolacon candezei* Fleutiaux, 1928, female, holotype. **1** habitus, dorsal view **2** ditto, ventral view **3** data labels.

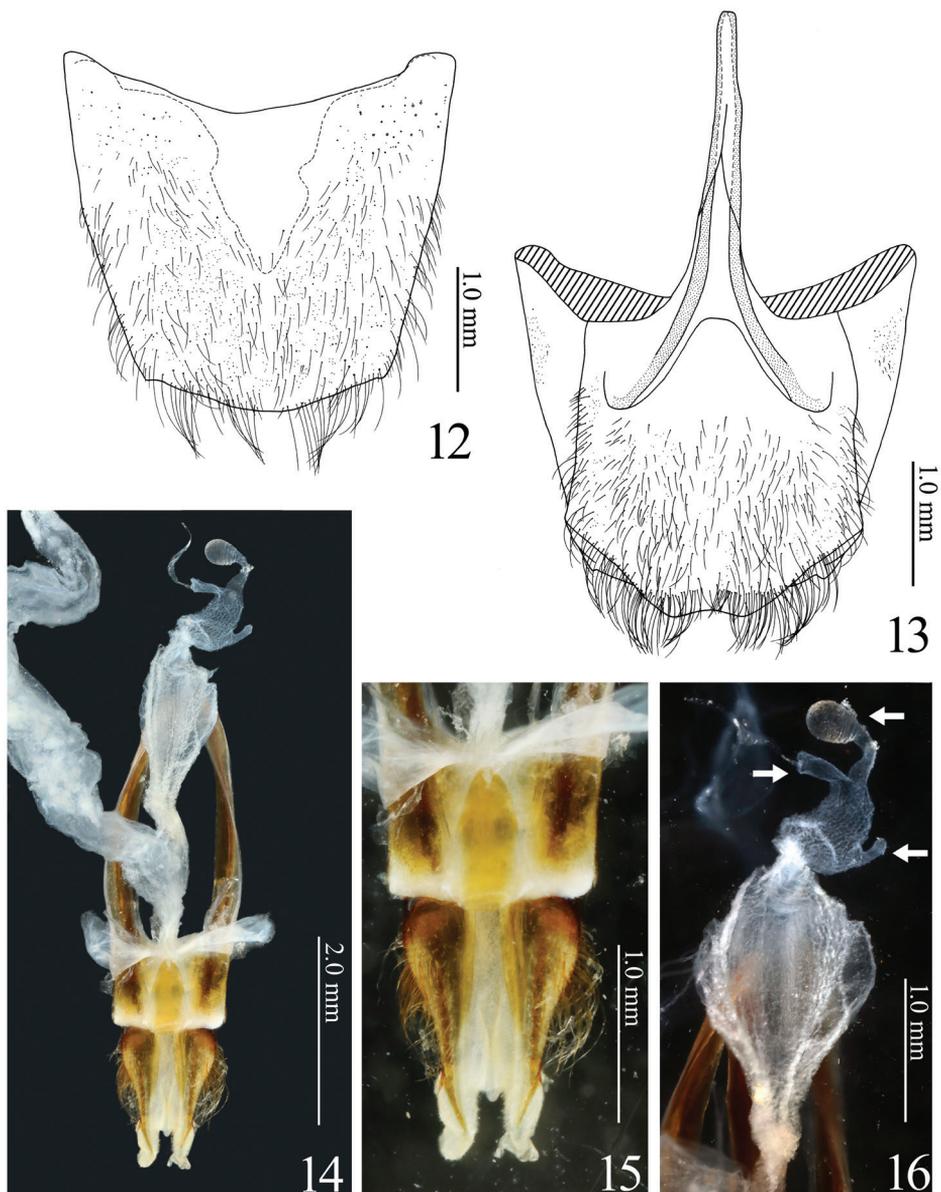
MP4 (Fig. 11: arrow); wedge cell absent. Elytra widest on basal half; intervals with uniformly small punctures.

Abdomen. Ventrite V 0.59 × as long as wide. Tergite VIII (Fig. 12) truncate apically. Sternite VIII (Fig. 13) widest at apical 1/3, apex concave; spiculum ventrale 1.4 × longer than sternite VIII.

Genitalia (Fig. 14). Ovipositor with coxites not sclerotized at apex (Fig. 15). Bursa copulatrix with three short sacs (Fig. 16: arrows); without sclerotized structures.



Figures 4–11. *Alaolacon candezei* Fleutiaux, 1928, female, holotype. **4** head and pronotum, anterolateral view **5** right antenna, anterior side (dotted line: apical half part of antennomere XI thinner than its basal half part) **6** mouth-parts **7** prosternal process, lateral view **8** anterior angle of hypomeron (arrow: mesal edge carinate anterolaterad) **9** posterior part of hypomeron and mesothorax, ventral view (arrows: posterior margin with two angles) **10** scutellum **11** right hind wing (arrow: cross vein between veins MP4 and CuA2 located at contact point between veins MP3 and MP4).



Figures 12–16. *Alaolacon candezei* Fleutiaux, 1928, female, holotype. **12** tergite VIII **13** sternite VIII, ventral view and tergite VIII, dorsal view **14** genitalia, dorsal view **15** coxites, dorsal view **16** vagina and bursa copulatrix, dorsal view (arrow: bursa copulatrix with three short sacs).

Male. Unknown.

Distribution. Malaysia: Sabah: Banggi Island.

Remarks. This species is similar to *Alaolacon cyanipennis* Candèze, 1865 in large body size (24.0 mm), black body and elytra with metallic luster, but is distinguished by

the following contrasting characters (*A. cyanipennis* in parentheses): female antennomere III pectinate (Fig. 5) (female antennomere III trapezoidal); prothorax widest posteriorly (Fig. 1) (prothorax widest at mid-length except for posterior angles, Fig. 17); scutellum widest near posterior 2/5 (Fig. 10) (scutellum near posterior 1/3); wedge cell of hind wings absent (Fig. 11) (wedge cell of hind wings present, Fig. 18); female sternite VIII with apex concave (Fig. 13) (female sternite VIII with apex rounded).

This species are known only from the female holotype. We predict that the males also exhibit blue elytra and with metallic luster, scutellum widest near posterior 2/5, hind wings without wedge cell and with cross vein between veins MP4 and CuA2 located at contact point between veins MP3 and MP4.

Alaolacon cyanipennis Candèze, 1865

Figures 17–19

Alaolacon cyanipennis Candèze, 1865: 13 (original description: type locality: Peninsular Malaysia); Gemminger and Harold 1869: 1498 (catalogue of Coleoptera); Candèze 1874: 114 (monograph); Candèze 1891: 29 (catalogue; description of type locality: Malacca); Schwarz 1906: 316 (catalogue); Hyslop 1921: 625 (type species); Schenkling 1925: 40 (catalogue); Fleutiaux 1926: 102 (catalogue); Casari-Chen 1993: (description; designation of homeotype); Suzuki 2004: 152 (record from Sumatra).

Type material. Lectotype. Female, Malacca, West Malaysia (Peninsular Malaysia), Malaysia, Janson coll. [BMNH] (Fig. 19). Label data: “Malacca”; [female symbol]; Janson coll/ 1903-130.; “Alaolacon/ cyanipennis/ Cdz. ”; “Alaolacon/ cyanipennis Cand./ Comp to RSNB/ smaller female/ C.M.F. von Hayek/ 1976”; “female int. genitalia/ delicate no plates/ C.M.F. von Hayek/ 1978”; “Antseps”; “mouthparts in/ separate vial/ C.M.F. von Hayek 1991/ by Casari-Chen”; and with the authors’ red lectotype label: “LECTOTYPE/ Alaolacon cyanipennis/ Candèze, 1865”.

Diagnosis. Body black, elytra blue-black and with metallic luster; setae white; female antennomere III trapezoidal, 1.4 × as long as wide; prothorax as long as wide, widest at mid-length except for posterior angles; pronotal anterior angles bisinuate and rounded; anterior angles of hypomeron rounded; prosternal spine inclined weakly behind procoxae; scutellum concave laterally, widest near posterior 1/3; hind wings with wedge cell, with cross vein between veins MP4 and CuA2 located at contact point between veins MP3 and MP4; female sternite VIII with rounded apex.

Description. See Casari-Chen (1993) for a detailed description.

Distribution. Malaysia: the Peninsular Malaysia. Indonesia: Sumatra.

Remarks. Candèze (1865) did not provide the number of the type specimens. Candèze (1865) mentioned that “*Elle a été découverte et apportée récemment en Europe par M. de Castelnau. Je l’ai vue dans sa collection, ainsi que dans celle de M. le comte de Mniszech*”. Mniszech’s collection went to Laporte de Castelnau, part of this went to



17



19



18

Figures 17–19. *Alaolacon cyanipennis* Candèze, 1865, female, lectotype **17** habitus, dorsal view **18** right hind wing **19** data labels and body parts.

Janson and then to BMNH. Candèze’s first collection of Elateridae (up to 1869) went to the BMNH, while a second collection of Elateridae went to IRSNB (Bousquet, 2016). BMNH can be most expected to hold types of this species because it was described before 1869. Label data of the examined specimen in BMNH agree with the original description. The external features of the specimen also agree with the original description. Thus, the specimen should be considered a syntype. Casari-Chen (1993) considered the type specimen as a homeotype. We designated the known syntype as lectotype to stabilize the classification.

We could not locate other syntypes including at IRSNB in this time. Laporte de Castelnau's first collection was given to the National Institution of the Promotion of Science in Washington DC but was probably destroyed by fire, while part of his later collection was left to the Melbourne Museum in Australia (Bousquet 2016).

Only female specimens are known (Candèze 1865; Suzuki 2004). Only this species exhibits hind wings with wedge cell in this genus, whereas the other species lost wedge cell of hind wings. We predict that the male could also be recognized by presence of the wedge cell.

***Alaolacon fujiokai* sp. n.**

<http://zoobank.org/FF52714A-2F8B-413C-9777-C96D283C3465>

Figures 20–37

Etymology. The name of this species honors Mr. Masahiro Fujioka for providing the material.

Type material. Holotype. Male, Tawau, East Malaysia (Sabah), Malaysia, V 2014 [ELKU].

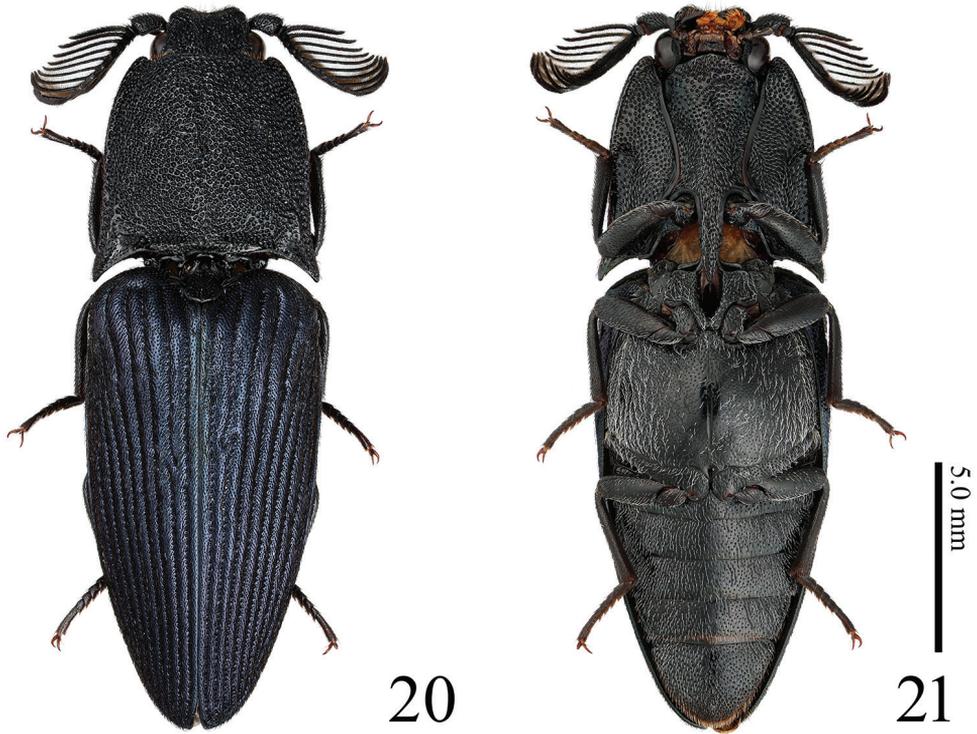
Diagnosis. Body black, elytra blue and with metallic luster, legs black; setae black on dorsal side and white on ventral side; frontal depression deep; eye small; prothorax almost as long as wide, widest posteriorly; pronotum with anterior angles bisinuate and rounded, median longitudinal depression deep, extending from before pronotal anterior margin to base, punctate; prosternal process inclined strongly behind procoxae; anterior angles of hypomerion acute; scutellum concave laterally, widest near posterior 1/3; hind wings with cross vein between veins MP4 and CuA2 located anterad to contact point between veins MP3 and MP4, without wedge cell; median lobe of male aedeagus stout.

Measurements. BL: 18.9, BW: 6.11, MIE: 2.08, MAE: 3.05, OI: 68, PL: 5.95, PML: 5.23; PW: 5.91, PI: 101, EL: 12.1, EW: 6.11, EI: 197.

Description of male. *Body* (Figs 20, 21) shiny, elytra with metallic luster. Color. Black except for elytra black-blue; mouth-parts brown-black, but mandible black, galea and lacinia orange; apical edge of tarsal segment V and tarsal claws red-brown; pregenital segments and aedeagus black-brown. Hairs. Dorsal surface covered with black flattened setae; ventral surface with white flattened setae; legs with intermixed black and white setae; mouth-parts and pronotal anterior margin near eyes with yellow-brown setae; filiform setae of antennomeres III–XII brown and long.

Head (Fig. 22). Frontal depression deep. Eyes small. Antennomere I long; antennomere II short, dish-shaped; antennomeres III–XI flabellation strong; antennomere XII elongate (Fig. 23). Apical maxillary palpomere 1.8 × as long as wide (Fig. 24) (Mandibles chipped in apical parts.)

Prothorax almost as long as wide, widest posteriorly; sides rounded anteriorly, linear posteriorly. Pronotum with anterior angles bisinuate and rounded; median longitudinal depression deep, extending from before pronotal anterior margin to base, punctate.



Figures 20, 21. *Alaolacon fujiokai* sp. n., male, holotype. **20** habitus, dorsal view **21** ditto, ventral view.

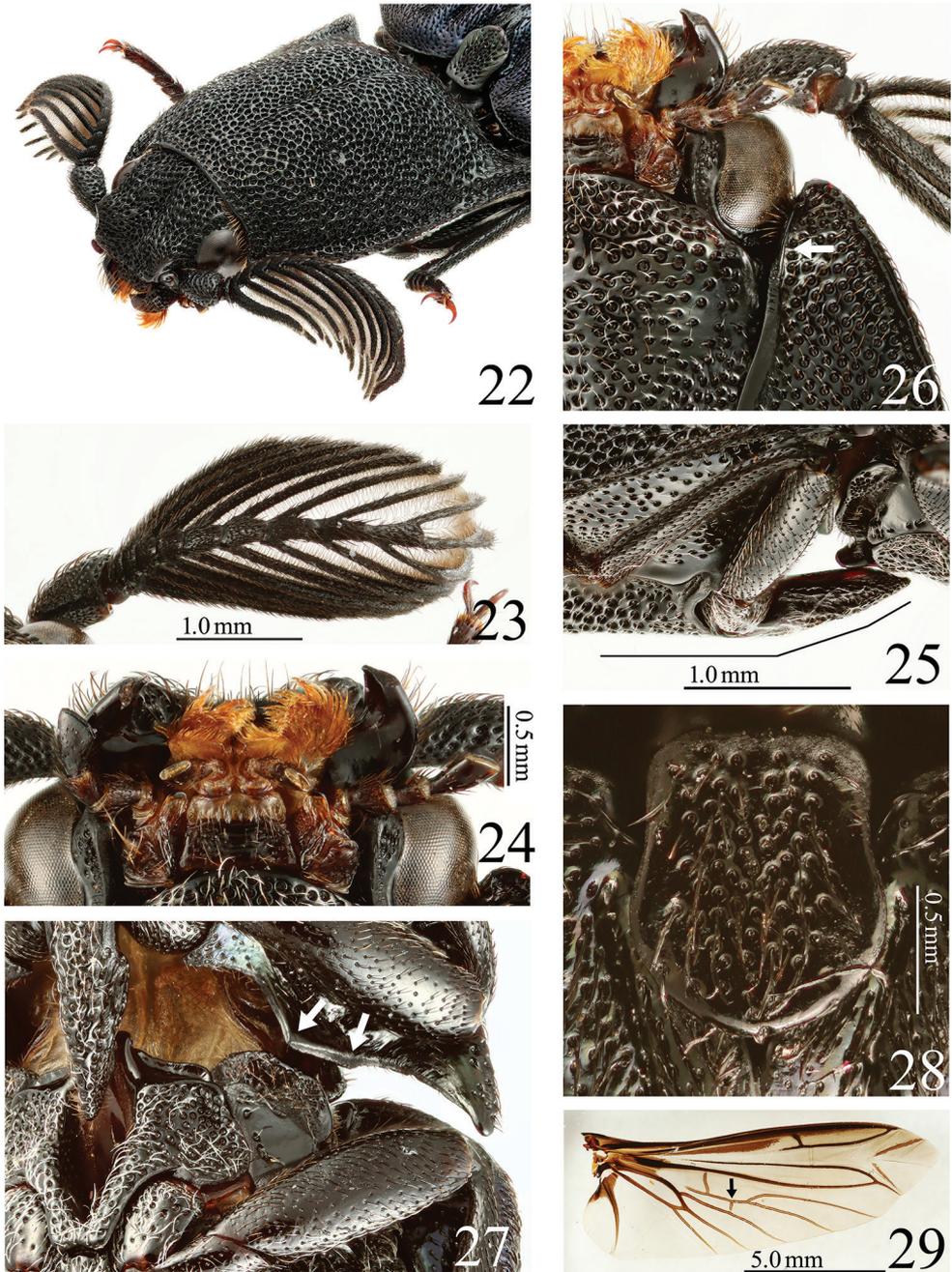
Prosternal spine inclined strongly (at 18 degrees) behind procoxae (Fig. 25). Hypomeron with anterior angles acute (Fig. 26). Scutellum (Fig. 28) concave laterally, $1.2 \times$ as long as wide, widest near posterior $1/3$. Hind wings with cross vein between veins MP4 and CuA2 apparent, not completely connected with CuA2, located anterad to contact point between veins MP3 and MP4 (Fig. 29: arrow); wedge cell absent. Elytra with sides almost parallel on basal half; intervals with small and coarse punctures.

Abdomen. Ventrite V $0.67 \times$ times as long as wide. Tergite VIII (Fig. 30) $0.72 \times$ as long as wide, colorless basal area. Sternite VIII (Fig. 31) with darker W-shaped band; median notch shallow and truncate transversally. Tergite IX (Fig. 32) with median notch shallow and rounded. Sternite IX (Fig. 33) narrowed abruptly on posterior half to apex. Aedeagus (Figs 34, 35). Median lobe stout; basal struts $0.35 \times$ total length of median lobe. Parameres with dense and long setae. Basal piece $0.29 \times$ total length of aedeagus.

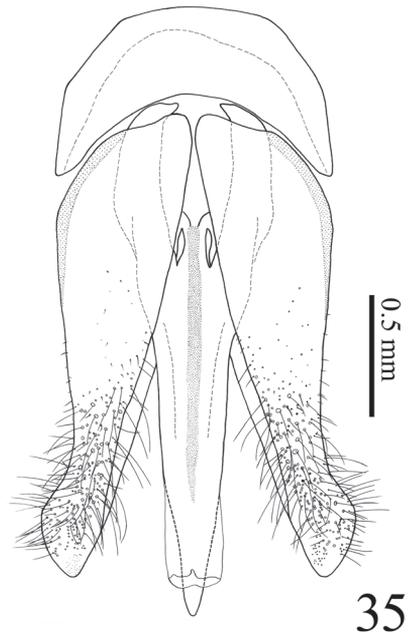
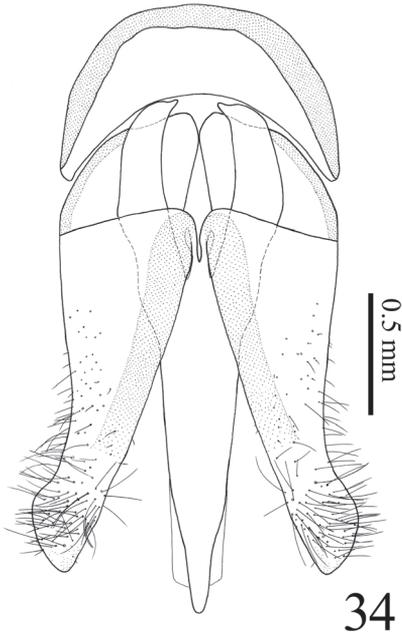
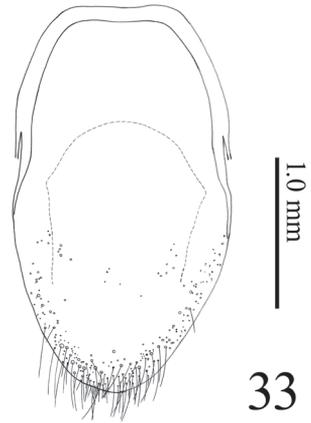
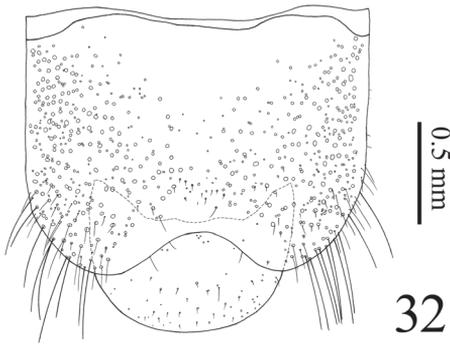
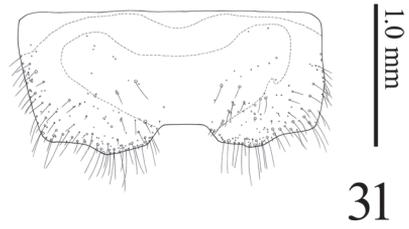
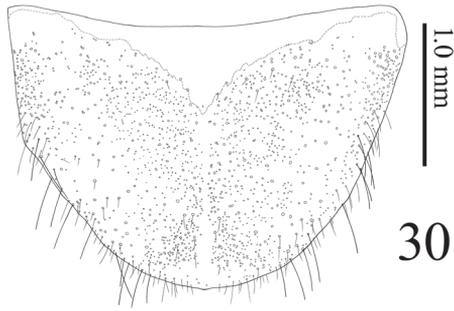
Female. Unknown.

Distribution. Malaysia: Sabah: Tawau.

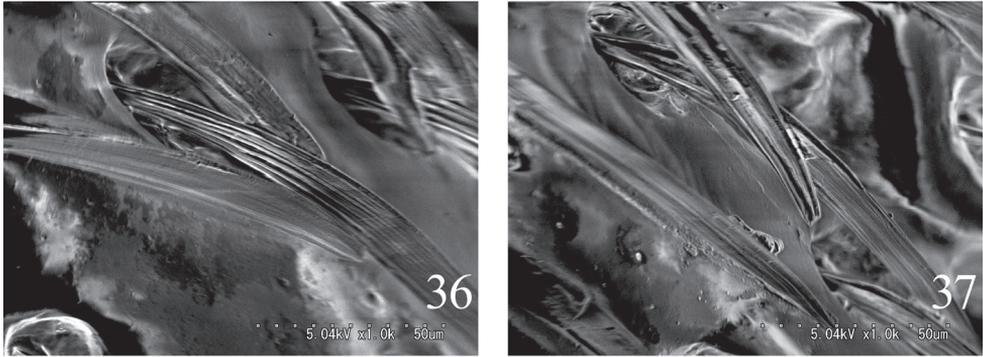
Remarks. This species is distinct by black body, blue elytra with metallic luster, black setae on dorsal side, white setae on ventral side and strong antennomeres III-XI flabellation. It is similar to *Alaolacon candezei* Fleutiaux, 1928 in having a black body, blue elytra with metallic luster, pronotum anterior angles bisinuate and rounded, and



Figures 22–29. *Alaolacon fujiokai* sp. n., male, holotype. **22** head and pronotum, anterolateral view **23** right antenna, dorsal view **24** mouth-parts **25** prosternal process, lateral view **26** anterior angle of hypomeron (arrow: mesal edge carinate anterolaterad) **27** posterior part of hypomeron and mesothorax, ventral view (arrows: posterior margin with two angles) **28** scutellum **29** right hind wing (arrow: cross vein between veins MP4 and CuA2 located anterad to contact point between veins MP3 and MP4).



Figures 30–35. *Alaolacon fujiokai* sp. n., male, holotype. **30** tergite VIII **31** sternite VIII **32** tergites IX–X **33** sternites IX–X **34** aedeagus, dorsal view **35** ditto, ventral view.



Figures 36, 37. *Alaolacon fujiokai* sp. n., male, holotype, setae of elytra. **36** median part **37** apical part.

scutellum concave laterally, except for drastic sexual differences of antennae, but is distinguished by the following contrasting characters (*A. candezei* in parentheses): legs black (Fig. 21) (legs red, Fig. 2); setae black on dorsal side and white on ventral side (Fig. 20) (all setae white, Fig. 1); frontal depression deep (Fig. 22) (frontal depression moderate, Fig. 4); pronotal median longitudinal depression extending from before pronotal anterior margin to base (Fig. 22) (pronotal median longitudinal depression not reaching anterior margin or base, Fig. 4); anterior angles of hypomeron acute (Fig. 26) (anterior angles of hypomeron rounded, Fig. 8); prosternal spin inclined strongly behind procoxae (Fig. 25) (prosternal spine inclined weakly behind procoxae, Fig. 7); scutellum widest near posterior 1/3 (Fig. 28) (scutellum widest near posterior 2/5, Fig. 10); hind wings with cross vein between veins MP4 and CuA2 located anterad to contact point between veins MP3 and MP4 (Fig. 29: arrow) (hind wings with cross vein between veins MP4 and CuA2 located at contact point between veins MP3 and MP4, Fig. 11: arrow).

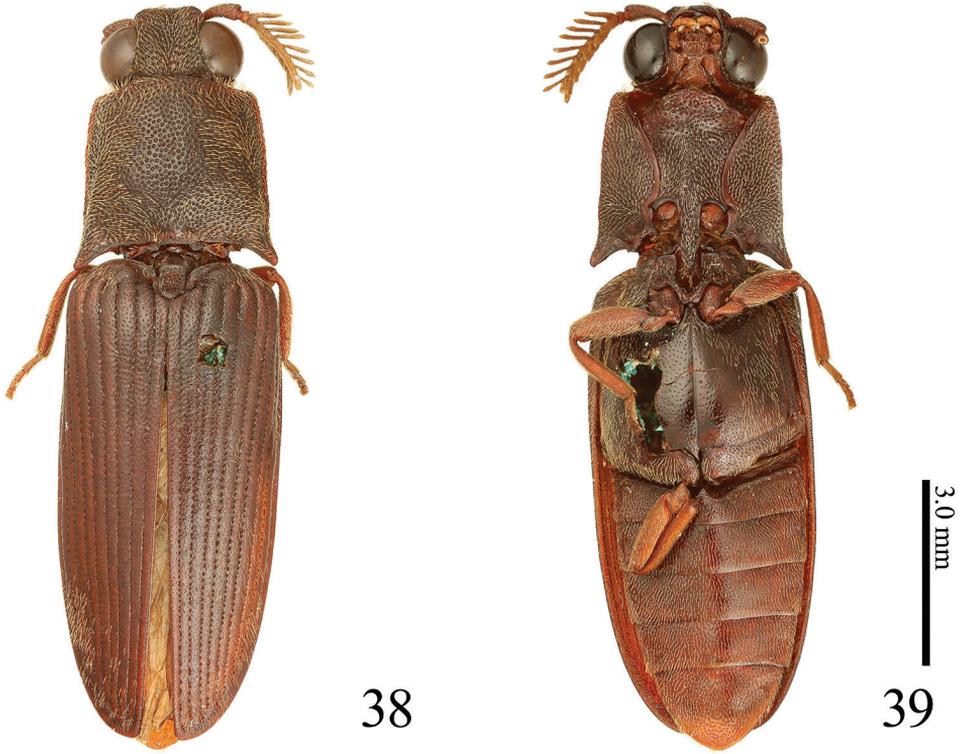
Alaolacon fujiokai and *A. candezei* are similar species from the same island, but we recognized they are different species by the setal color and the hind wing venation. We believe that setal complementary color difference probably is not caused by sexual dimorphism because such dimorphism has not previously been observed in species of the Agrypninae. We also believe that differences in hind wing venation are unlikely to be caused by sexual dimorphism because such dimorphism has not previously been observed in species with flying females.

Alaolacon megalopus sp. n.

<http://zoobank.org/83188584-DF58-41F6-BF06-BF6702F909A8>

Figures 38–54

Eumoes murrayi Candèze, 1874; Fleutiaux 1928: 178 (mention a specimen from Java at IRSNB); Casari-Chen 1993: 241 (examined a male specimen from Java). Misidentification.



40

Figures 38–40. *Alaolacon megalopus* sp. n., male, holotype. **38** habitus, dorsal view **39** ditto, ventral view **40** data labels.

Etymology. A combination of the Greek *megalos*, meaning great, and the Greek *ops*, meaning eye, refer to the large compound eyes.

Type material. Holotype. Male, Java, Indonesia [IRSNB] (Fig. 40).

Diagnosis. Body brown, without metallic luster; setae yellow-brown; frontal depression moderate; eye very large; prothorax wider than long, widest posteriorly; pronotal anterior angles rounded; median longitudinal depression shallow, located at

anterior half, punctate; anterior angles of hypomerion acute; prosternal spine inclined strongly behind procoxae; scutellum $1.5 \times$ as long as wide; sides of scutellum parallel; hind wings with cross vein between veins MP4 and CuA2 located just anterior to contact point between veins MP3 and MP4, without wedge cell; male tergite VIII longer than wide; median lobe of male aedeagus elongate.

Measurements. BL: 11.8, BW: 3.54, MIE: 1.02, MAE: 2.31, OI: 44, PL: 3.11, PML: 2.74, PW: 3.26, PI: 95, EL: 7.70, EW: 3.54, EI: 218.

Description. *Body* (Figs 38, 39) shining, without metallic luster. Color. Body brown; antennomere I, pronotal lateral margin, elytra, legs, abdomen paler; antennomeres II–XII, mouth-parts, pregenital segments and aedeagus yellow-brown, but mandible brown. Hairs. Body covered with yellow-brown setae; antennomeres III–XII with short filiform setae at ventral surface.

Head (Fig. 41). Frontal depressed moderate. Eyes very large. Antennomere I elongate; antennomere II short and obconical; antennomeres III–X flabellation moderate (Fig. 42). Apical maxillary palpomere (Fig. 43) rounded, $1.3 \times$ as long as wide. (Antennomeres XI–XII of right antenna and antennomeres III–XII of left antenna lost.)

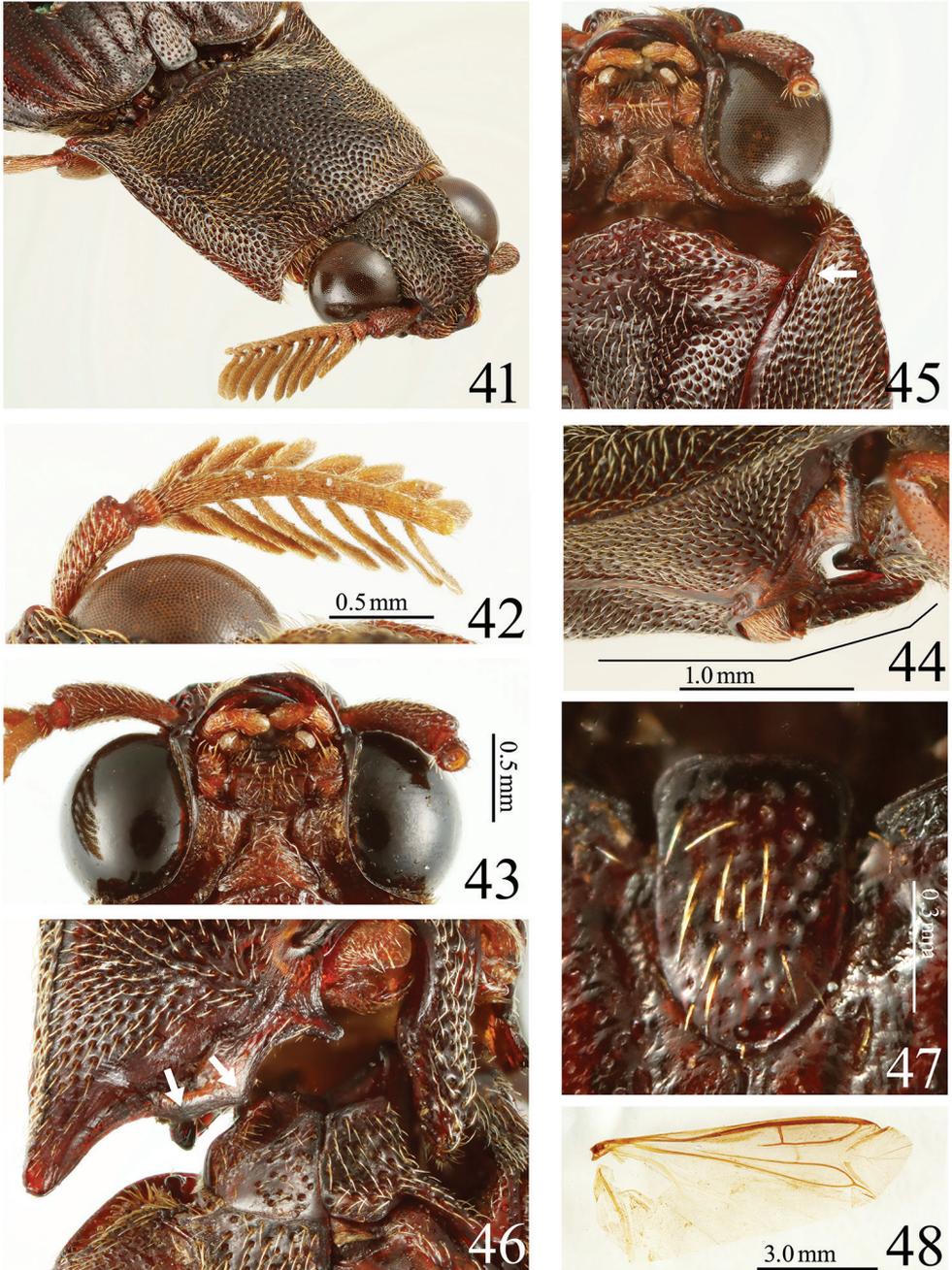
Prothorax wider than long; sides widest posteriorly, rounded anteriorly, lined posteriorly. Pronotum convex; anterior angles rounded; median longitudinal depression shallow, located at anterior half, punctate (Fig. 41); central area with two shallow concaves. Prosternal spine with lateral margin of dorsal side expanded laterally, inclined strongly (at 15 degrees) behind procoxae (Fig. 44). Hypomerion with anterior angles acute (Fig. 45); punctures more homogeneous than prosternal punctures in density and size. Scutellum (Fig. 47) $1.5 \times$ as long as than wide; sides parallel. Elytra with sides almost parallel on basal half; intervals with small and coarse punctures. Hind wings (Fig. 48) with veins posterior to MP3 translucent; cross vein between veins MP4 and CuA2 not completely connected with CuA2, located just anterior to contact point between veins MP3 and MP4; wedge cell absent (Fore legs except for coxae, tarsomeres IV–V and claw of right middle leg, tarsomere V and claw of left middle leg, tarsi and claw of right hind leg, and left hind leg lost.)

Abdomen. Ventrite V $0.65 \times$ as long as wide. Tergite VIII (Fig. 49) $1.2 \times$ as long as wide; basal area translucent. Sternite VIII (Fig. 50) with central area translucent; median notch shallow and truncate transversally. Tergite IX (Fig. 51) with median notch shallow and rounded. Tergite X (Fig. 51) posterior margin rounded. Sternite IX (Fig. 52) wide; posterior half abruptly narrowed to apex; posterior margin rounded. Aedeagus (Figs 53, 54). Median lobe elongate, basal struts $0.37 \times$ total length of median lobe. Parameres with sparse and short setae. Basal piece $0.28 \times$ total length of aedeagus.

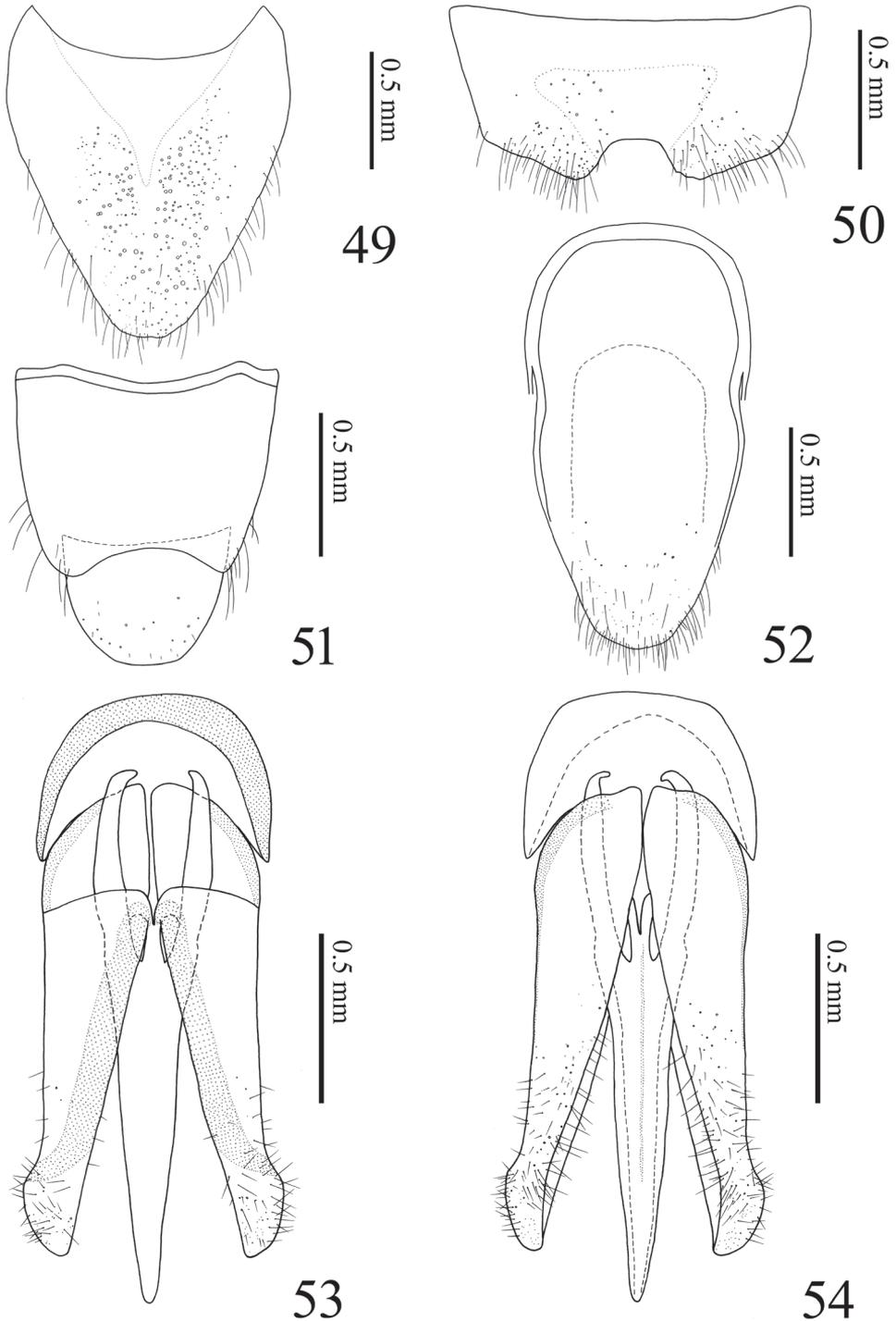
Female. Unknown.

Distribution. Indonesia: Java.

Remarks. The holotype is damaged with most appendages lost. The holotype of this species was identified as *Eumoeus murrayi* (= *Alaolacon murrayi* comb. n.) by Candèze (Fleutiaux, 1928), but separated from *A. murrayi* by the following characteristics (the holotype of *A. murrayi* in parentheses): eye very large (OI: 44) (eye large, OI: 50); anterior angles of pronotum rounded (Fig. 38) (anterior angles of pronotum bisinuate,



Figures 41–48. *Alaolacon megalopus* sp. n., male, holotype. **41** head and pronotum, anterolateral view **42** right antenna, dorsal view **43** mouth-parts **44** prosternal process, lateral view **45** anterior angle of hypomeron (arrow: mesal edge carinate anterolaterad) **46** posterior part of hypomeron and mesothorax, ventral view (arrows: posterior margin with two angles) **47** scutellum **48** right hind wing.



Figures 49–54. *Alaolacon megalopus* sp. n., male, holotype. **49** tergite VIII **50** sternite VIII **51** tergites IX–X **52** sternites IX–X **53** aedeagus, dorsal view **54** aedeagus, ventral view.

Fig. 56); pronotal median longitudinal depression shallow, located at pronotal anterior half and punctate (Fig. 41) (pronotal median longitudinal depression not reaching anterior margin or base and impunctate at posterior half); scutellum $1.5 \times$ as long as wide (Fig. 47) (scutellum $1.3 \times$ as long as wide, Fig. 56); scutellum sides parallel (Fig. 47) (scutellum sides concave and widest posteriorly, Fig. 56); hind wings with cross vein between veins MP4 and CuA2 (Fig. 48) (hind wings without cross vein between veins MP4 and CuA2); male tergite VIII longer than wide (Fig. 49) (male tergite VIII shorter than wide).

Only this species exhibits parallel sides of scutellum in this genus. The scutellum shape could be a useful specific diagnostic feature for this species including its female.

***Alaolacon murrayi* (Candèze, 1874), comb. n.**

Figures 55–58

Eumoeus murrayi Candèze, 1874: 113 (original description on male; type locality: Madras, India), 214 (as “*Eumaeus murrayi*”; index); Schwarz 1906: 40 (catalogue); Hyslop 1921: 645 (type species); Schenkling 1925: 51 (as “*Eumaeus*”; catalogue); Fleutiaux 1928: 178 (comments); Casari-Chen 1993: 241 (description on male; examination of holotype; misspelled *E. murrayi*); Casari 2008: 158 (morphological phylogeny of Hemirhipini genera; misspelled *E. murrayi*), 161 (drawing of habitus).

Tharopsides harmandi Fleutiaux, 1918: 235 (original description on male; type locality: Bangkok, Thailand); Fleutiaux 1924: 177 (republish of original description); Schenkling 1927: 509 (catalogue of world Elateridae); Fleutiaux 1940: 40 (record of male from Vietnam); Fleutiaux 1947: 307 (as synonymy of *Luzonicus murrayi* (Candèze, 1874)).

Luzonicus murrayi (Candèze, 1874): Fleutiaux 1947: 307 (change generic status; description).

Type material. Holotype. Male, Madras, India, Murray leg. [IRSNB] (Fig. 58).

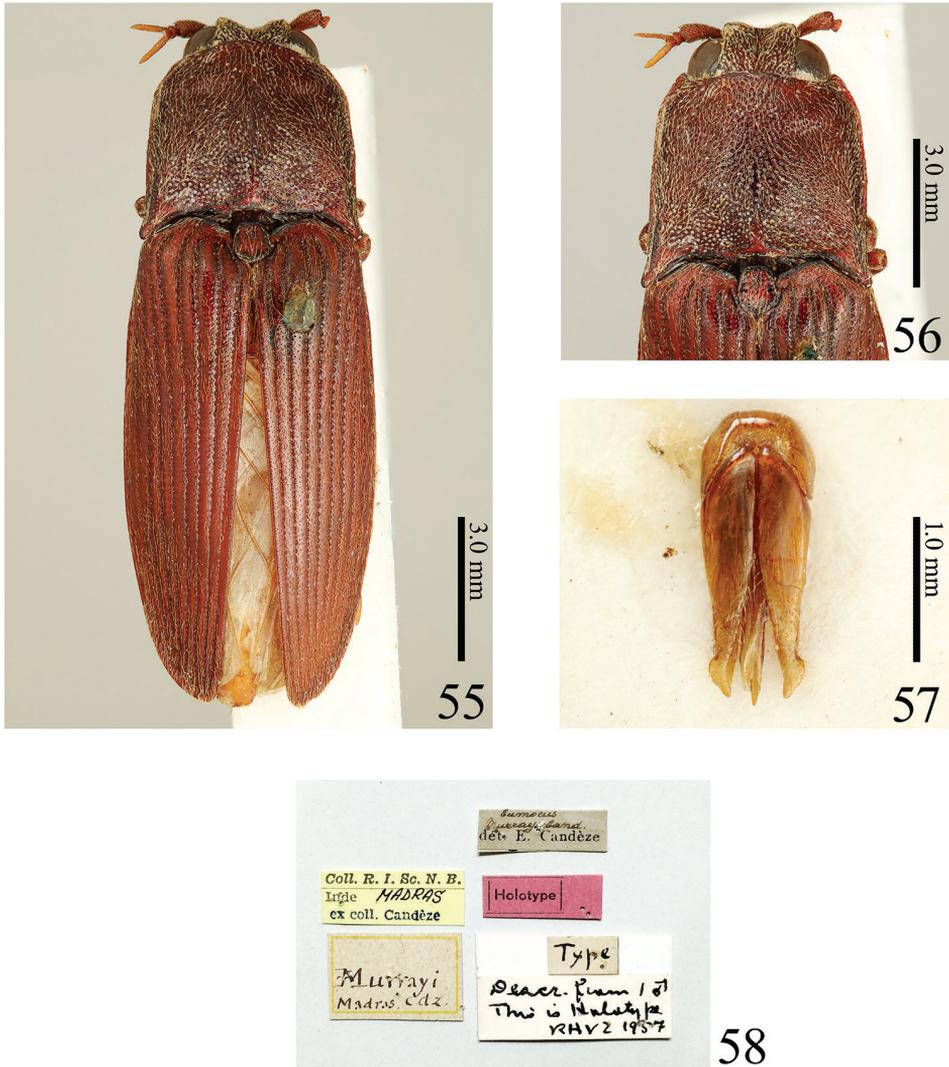
Diagnosis. Body red-brown, without metallic luster; setae yellow-brown; frontal depression deep; eye large; prothorax shorter than wide, widest posteriorly; pronotum with anterior angles bisinuate, median longitudinal depression not reaching anterior margin or base and impunctate at posterior half; anterior angles of hypomeron acute; prosternal spine inclined strongly behind procoxae; scutellum $1.3 \times$ as long as wide, with sides straight, widest posteriorly; hind wings without cross vein between veins MP4 and CuA2 and wedge cell; male tergite VIII shorter than wide; median lobe of male aedeagus elongate.

Measurements. BL: 14.9, BW: 4.85, MIE: 1.43, MAE: 2.85, OI: 50, PL: 4.34, PML: 3.68, PW: 4.64, PI: 94, EL: 10.1, EW: 4.85, EI: 208.

Description. See Casari-Chen (1993) for a detailed description.

Distribution. India. Thailand. Vietnam.

Remarks. This species is only known from the male.



Figures 55–58. *Alaolacon murrayi* (Candèze, 1874), comb. n., male, holotype. **55** habitus, dorsal view **56** head, pronotum and scutellum **57** aedeagus, ventral view **58** data labels.

Discussion

Candèze (1874) produced a misspelling of *Eumoeus*, writing “EUMÆUS” in the index on page 214. Candèze (1891) used “EUCEUS” on page 29 and “Eumœus” in index on page 243. This means that Candèze (1891) had already recognized *Eumoeus* as a valid name. However, Fleutiaux (1940) considered *Eumoeus* as a wrong spelling of *Eumaeus* and used *Eumaeus* as the valid name. He used *Tharopsides* Fleutiaux, 1918 as the replacement name for “*Eumaeus* Candèze, 1874” because it became a junior homonym for the genus *Eumaeus* Hubner, 1816 of Lepidoptera.

Eumoeus and *Tharopsides* were described from males exhibiting 12-segmented and biflabellate antennae, whereas *Luzonicus* were described from female exhibiting 11-segmented and filiform to subpectinate antennae. Fleutiaux (1947) inferred that there was an occurrence of sexually dimorphic antennae of these genera, and that *Luzonicus* was therefore the senior synonym for *Eumoeus* and *Tharopsides*. Actually *Eumoeus* is the senior synonym for both *Luzonicus* and *Tharopsides* because the actions of Fleutiaux (1940) are nullified.

Alaolacon Candèze, 1865 was only known from female with 11-segmented and pectinate antennae. We determined that a male specimen (the holotype of *A. fujiokai* sp. n.), in possessing biflabellate antennae, belongs to *Alaolacon* because of the similarity to *Alaolacon cyanipennis* and *Alaolacon candezei* including: black body, blue elytra with metallic luster, pronotum anterior angles bisinuate, scutellum concave laterally. This association demonstrates that *Alaolacon* also has sexually dimorphic antennae.

In the tribe Hemirhipini, only four genera, *Alaolacon*, *Eumoeus*, *Mocquersysia* Fleutiaux, 1899 and *Eleuphemus* Hyslop, 1921 have strongly sexually dimorphic antennae. Their males exhibit 12-segmented and biflabellate antenna, and females exhibit 11-segmented and subpectinate antennae. *Eleuphemus* is separated from *Alaolacon*, *Eumoeus*, *Mocquersysia* (the latter three genera in parentheses) by the supra-antennal carinae continuous across frons (supra-antennal carina not continuous across frons) and mandible without subapical tooth (mandible with subapical tooth). *Mocquersysia* is separated from *Alaolacon* and *Eumoeus* (the latter two genera in parentheses), prosternal suture shortly grooved (prosternal suture not grooved), scutellum narrowed apically and with straight side (scutellum widest apically and concave laterally or with parallel sides in *A. megalopus* sp. n.), elytral intervals flat (elytral intervals convex).

Candèze (1874) suggested that *Alaolacon* should be combined with *Eumoeus* if its male had biflabellate antennae. We recognized that *Alaolacon* and *Eumoeus* are similar by many structures: setae flat, wider at midlength than base, with longitudinal microcarinae (Figs 36, 37); interspaces greater than puncture diameter except for smaller on head and pronotum; hypomerion mesal edge carinate anterolaterad (Figs 8, 26, 45: arrow); hind wings with vein r4 translucent (Figs 11, 18, 29, 48). The two genera could not be separated from each other except by antennal morphology. This non-antennal similarity suggests that the two genera should be considered synonyms because antennal morphology is dimorphic in several other Elateridae. We propose that the two genera should be considered synonyms. Accordingly, the priorities of the generic names are following: *Tharopsides* < *Luzonicus* < *Eumoeus* < *Alaolacon*.

Luzonicus bakeri Fleutiaux, 1916 and *T. bakeri* Fleutiaux, 1918 are eventual homonyms since *Luzonicus* and *Tharopsides* are junior synonyms of *Alaolacon*. We propose *A. philippinensis* nom. n., as the replacement name for *A. bakeri* (Fleutiaux, 1916) comb. n. *Alaolacon* currently contains eight species, 1, *A. bakeri*, 2, *A. candezei*, 3, *A. cyanipennis*, 4, *A. fujiokai*, 5, *A. griseus* Candèze, 1874, 6, *A. megalopus*, 7, *A. murrayi* and 8, *A. philippinensis*.

We could not find the types of two species, *A. bakeri* and *A. philippinensis*, and have not examined these species. Further effort to find the localities of the types of the two species are needed in order to understand the complete picture of these species.

Key to species for adults of the genus *Alaolacon*

- 1 Head and pronotum brown to red-brown.....2
 – Head and pronotum black.....4
 2 Prothorax longer than wide, elytra red-brown but brown-black on posterior half*A. bakeri* (Fleutiaux, 1916)
 – Prothorax shorter than wide, elytra brown to red-brown.....3
 3 Scutellum widest posteriorly*A. murrayi* (Candèze, 1874) **comb. n.**
 – Scutellum with parallel sides*A. megalopus* sp. n.
 4 Elytra blue or blue-black, and with metallic luster5
 – Elytra black and without metallic luster7
 5 Setae black dorsally and white ventrally*A. fujiokai* sp. n.
 – All setae white.....6
 6 Prothorax widest posteriorly, wedge cell of hind wings absent.....
*A. candezei* Fleutiaux, 1928
 – Prothorax widest at mid-length except for posterior angles, wedge cell of hind wings present*A. cyanipennis* Candèze, 1865
 7 Ventral surface red-brown.....*A. griseus* Candèze, 1874
 – Ventral surface black.....*A. philippinensis* nom. n.

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Pseudomacrochenus wusuae sp. n., a new species from Sichuan, China (Coleoptera, Cerambycidae, Lamiinae)

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Abstract

Pseudomacrochenus wusuae sp. n. (Coleoptera, Cerambycidae, Lamiinae, Lamiini) is described from Sichuan, China. Relevant morphological characters are illustrated by colour plates and a differential diagnosis of the new species from its relatives is provided.

Keywords

Cerambycidae, China, Lamiinae, Lamiini, new species, *Pseudomacrochenus*, taxonomy

Introduction

The Oriental genus *Pseudomacrochenus*, belonging to the tribe Lamiini in the subfamily Lamiinae (Coleoptera: Cerambycidae), was originally established by Breuning (1943), with *Pelargoderus antennatus* Gahan, 1894 as the type species fixed by the original designation.

Pseudomacrochenus Breuning, 1943 is a small genus only composed of five valid species: *P. antennatus* (Gahan, 1894), *P. spinicollis* Breuning, 1949, *P. oberthueri* Breuning, 1955, *P. affinis* Breuning, 1960, *P. albipennis* Chiang, 1981; their geographical distributions are generally limited to the northern Oriental region. Undoubtedly, China is the distribution center of *Pseudomacrochenus* since four species were recorded and two of them are endemic to China (Wang 1997, Löbl & Smetana 2010). In this paper, a new species is described: *P. wusuae* sp. n., which was collected from Xichang City, Sichuan Province, China.

Material and methods

Specimens were relaxed and softened in a hot saturated solution of potassium hydroxide for three minutes, and then transferred into distilled water to rinse the residual potassium hydroxide off and stop any further bleaching. The softened specimens were moved into glycerin and dissected there to observe morphological details. After examination, the body parts were mounted on plastic slips with gum arabic for future studies. Habitus photographs were taken using a Canon 50D DSLR with EF 100mm f/2.8L IS USM lens. Observations, photographs and measurements of morphological details were performed using a Zeiss Axio Zoom V16 motorized stereo zoom microscope (magnification up to $\times 270$) with a Zeiss AxioCam MRc 5. The final deep focus images were created with Helicon Focus 5.3 or Zerene Stacker 1.04 stacking softwares. Adobe Photoshop® CS6 was used for post processing. Measurements are averaged over five specimens.

Relevant morphological characters are illustrated with colour plates and a differential diagnosis of the new species from its relatives is provided.

The material examined for this study is deposited in the following collections and museums:

BITS	Bin Insect Taxonomy Studio, Beijing, China
CCWI	Collection of Wen-I Chou, Taitung, Taiwan, China
CCZC	Collection of Chao Zhou, Chengdu, Sichuan, China
CJYT	Collection of Junsuke Yamasako, Tokyo, Japan
CLHC	Collection of Li He, Chengdu, Sichuan, China
NMPC	Národní museum, Prague, Czech Republic

Results

Genus *Pseudomacrochenus* Breuning, 1943

Vernacular name: 伪鹿天牛属

Distribution. North Oriental.

***Pseudomacrochenus wusuae* sp. n.**

<http://zoobank.org/C9924F5B-C4EE-4B95-BD8F-324CCA00FF89>

Vernacular name: 午苏伪鹿天牛

Figs 1, 2, 3, 4B, F–H, 5, 6C–D

Type material. Holotype: ♂, CHINA, Sichuan: Liangshan Yi Autonomous Prefecture, Xichang City, Mt. Lushan (泸山) N27°49', E102°15', alt. 2050 m, 8.V.2015, Li He leg. (BITS); **Paratype:** 23♂♂39♀♀. 5♂♂4♀♀, same data as holotype except 16–17.XI.2015 (larva), em. II–III.2016, Li He & Bin Liu leg. (5♂♂ in NMPC and 4♀♀ in CLHC); 2♀♀, same data as holotype except alt. 2230 m, 8.VIII.2012, Li He leg. (CLHC); 1♀, same data as holotype (CLHC); 1♀, same data as holotype except 16–17.XI.2015 (larva), 17.II.2016 (pupa), em. 16.III.2016, Li He & Bin Liu leg. (CLHC); 1♀, same data as holotype except 16.XI.2015 (larva), em. 3.III.2016, Li He & Bin Liu leg. (BITS); 1♂, same data as holotype except alt. 2095 m, 17.XI.2015 (larva), em. 16.II.2016, Li He & Bin Liu leg. (BITS); 2♀♀, same data as holotype except 5.III.2016 (larva), em. 3.IV.2016, Li He leg. (CLHC); 7♂♂7♀♀, same data as holotype except 7.V.2016, Li He & Ben-Fu Miao leg. (CLHC); 1♂, same data as holotype except alt. 1928 m, 11.VI.2016, Bin Liu leg. (BITS); 2♂♂7♀♀, same data as holotype except alt. 1900 m, 11–16.VI.2016, Bin Liu leg. (CJYT); 1♂, same data as holotype except alt. ca. 2000 m, 12.VI.2015, Bin Liu leg. (CJYT); 1♂1♀, same data as holotype except alt. 1900 m, 11–16.VI.2016, Bin Liu leg. (CCWI); 1♂2♀♀, same data as holotype except alt. 1928 m, 12.VI.2016, Bin Liu leg. (BITS); 2♀♀, same data as holotype except alt. 2040 m, 12.VI.2016, Bin Liu leg. (BITS); 1♀, same data as holotype except alt. 1928 m, 13.VI.2016, Bin Liu leg. (BITS); 5♀♀, same data as holotype except alt. 2049 m, 16.VI.2016, Bin Liu leg. (BITS); 1♂, same data as holotype except alt. 1703 m, 20.VI.2016, Bin Liu leg. (BITS); 1♀, same data as holotype except alt. 1928 m, 20.VI.2016, Bin Liu leg. (BITS); 1♀, same data as holotype except alt. 1647 m, 21.VI.2016, Bin Liu leg. (BITS); 1♂1♀, same data as holotype except 25.VI.2016, Chao Zhou, Bin Liu & Li He leg. (CCZC); 1♂, same data as holotype except 25.VI.2016, Chao Zhou, Bin Liu & Li He leg. (BITS).

Diagnosis. Pronotum without spine at the lateral side, but only with an inconspicuous vestigial small tubercle. Elytra with a contrasting large spot on the middle constituted of pale grayish setae, except for a hairless area around the anterior margin forming a black semicircular ring. Abdominal tergite VIII with posterior edge weakly emarginate; sternite VIII short, with posterior edge more or less truncate.

Description. Male. Size relatively large, body length 16.22–30.20 mm, humeral width 4.48–8.72 mm. Length (mm) of different body parts: head (3.53) : antenna (68.27) : pronotum (5.37) : elytra (17.39) : protibia (7.57); width (mm): head (3.44) : pronotum (4.91) : elytra (7.84). Body length/elytral width = 3.50; antenna length/body length = 2.49. Antennomeres with length ratio from base to tip: 6.70 - 1.00 - 16.38 - 12.29 - 12.33 - 12.80 - 12.92 - 11.78 - 9.70 - 7.85 - 13.64.

Habitus is shown in Fig. 1A–B. Body color dark brown to black. Head covered with fulvous setae, forming four small spots at posterior margin of the occiput.

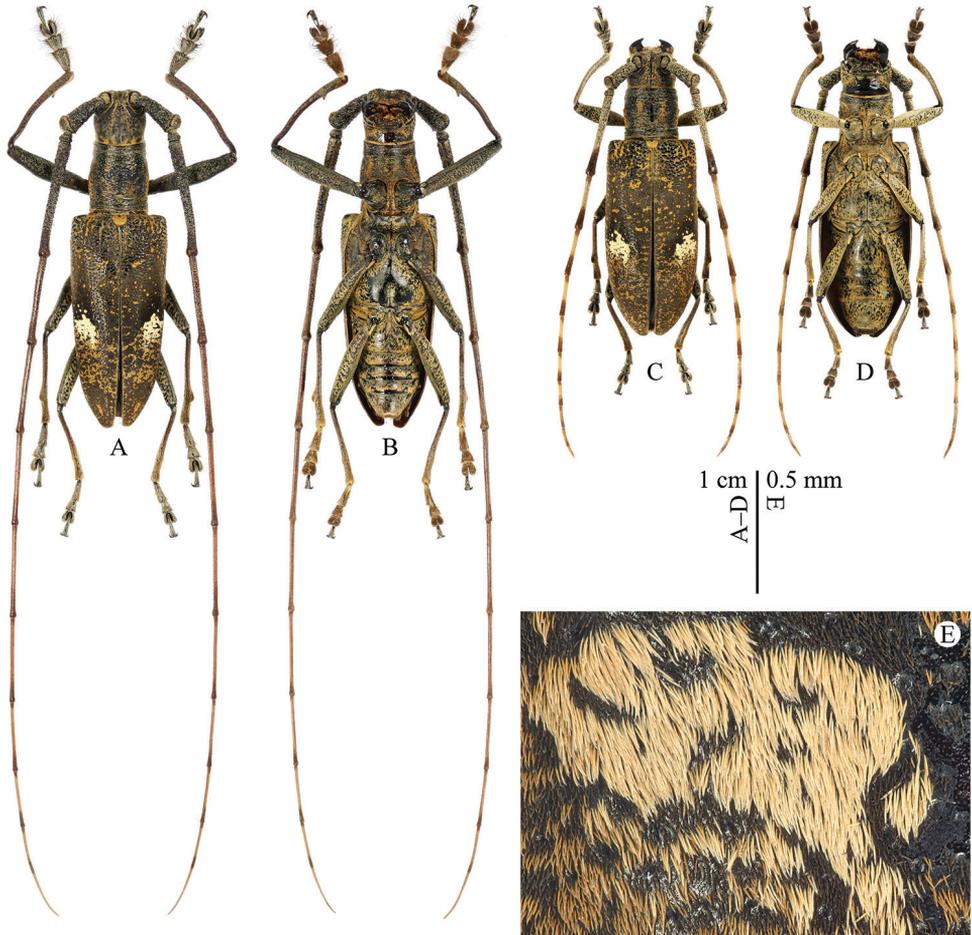


Figure 1. *Pseudomacrochenus wusuae* sp. n. **A** ♂ habitus (holotype; dorsal view) **B** ♂ habitus (holotype; ventral view) **C** ♀ (paratype; dorsal view) **D** ♀ (paratype; ventral view) **E** magnification of a grayish large spot on the elytron (♂; paratype; dorsal view).

Dorsal surface of scape, pedicel and antennomere III with fulvous setae, following antennomeres with very faint fulvous setae; apical parts of the antennomeres III–X and middle part of the antennomere XI with very faint brown setae, making alternating contrasting colors on these antennomeres; ventral surface of scape, pedicel and antennomeres III–IV fringed with long, brown setae, but distinctly less dense on the antennomere IV. Pronotum with middle line flanked by two ill-defined longitudinal fasciae of fulvous setae and each pronotal side with another one. Elytra mostly covered with very faint brown setae; a contrasting large spot on the middle constituted of pale grayish setae (Fig. 1E), except for a hairless area around the anterior margin forming a black semicircular ring; a number of irregularly scattered small spots of fulvous setae forming three or four short longitudinal fasciae contiguous at base and much denser and contiguous after the discal spot. Variations of pubescence



Figure 2. Variations of pubescence and spots on the elytra of *Pseudomacrochenus wusuae* sp. n. (paratypes; dorsal view) **A–D** ♂♂, **E–H** ♀♀.

is shown in Fig. 2A–D. Abdomen covered with fulvous setae, lateral margins with some erected brown setae.

Head (Fig. 3A) compressed, surface coarsely granulated, length/width = 1.03, narrower than pronotum. Frons transverse, slightly convex. Eyes small, finely faceted, divided into two widely separated lobes; lower lobes longer than genae. Interantennal region strongly concave between the strongly elevated antennal tubercles. Antennae long, extending beyond elytral apex by six antennomeres; scape stout and cylindrical, larger at apex, with an open circular scar; surface of scape, pedicel and antennomere III with small and coarse granules; antennomere III longer than all others; antennomeres IV, V, VI, VII and XI subequal; antennomeres VII to X decreasing in length; last antennomere thinner and curved.

Pronotum (Fig. 3C) slightly convex and elongate, length/width = 1.09, widest at about basal 3/7 where it is slightly protruded; anterior margin slightly concave; lateral

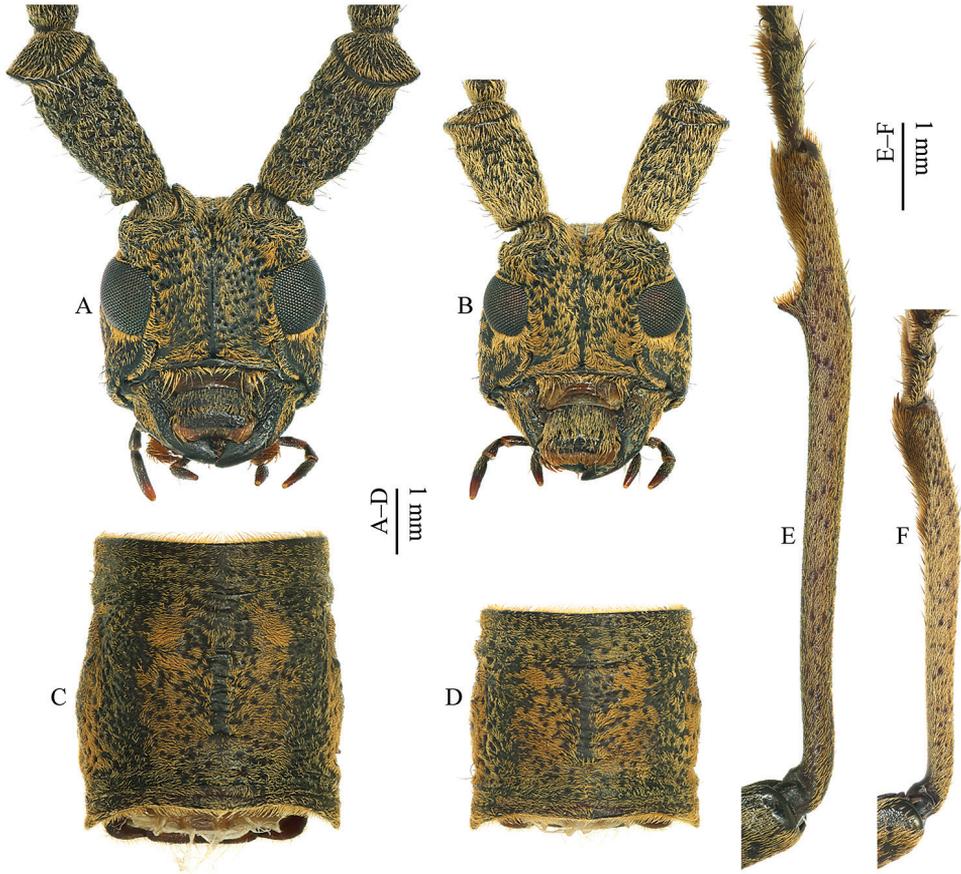


Figure 3. *Pseudomacrochenus wusuae* sp. n. (paratype). **A** head (♂; front view) **B** head (♀; front view) **C** pronotum (♂; dorsal view) **D** pronotum (♀; dorsal view) **E** protibia (♂; dorsal view) **F** protibia (♀; dorsal view).

side with an inconspicuous vestigial small tubercle; hind angles slightly projected backwards and somewhat acute; surface coarsely rugose.

Stridulatory organ hidden with a median longitudinal band of dense, fine, transverse stridulatory striae.

Scutellum ligulate, surface densely covered with fulvous setae.

Elytra widest just after humeri, length/width = 2.22, gradually narrowing towards apex; apices narrowly rounded; surface with many small and coarse granules at base, more or less thickly punctured with punctures diminishing in size towards apex.

Metathoracic wings fully developed.

Prolegs elongated; protibia (Fig. 3E) slightly sinuate, with a strong tooth at about the apical fourth of inner side. Profemora longer than meso- and metafemora. Metafemora exceed the posterior edge of visible abdominal segment IV.

Abdomen (Fig. 4B): tergite VIII with posterior edge weakly emarginate, bordered with long setae; sternite VIII short, with posterior edge more or less truncate, bordered with much shorter setae; sternite IX 'Y'-shaped.



Figure 4. *Pseudomacrochenus* species. ♂♂. **A, C–E** *P. antennatus* (Gahan, 1894) (Yunnan) **B, F–H** *P. wusuae* sp. n. (paratype) **A–B** tergites VIII, sternites VIII & IX (ventral view) **C–H** male genitalia (**C, F** dorsal view **D, G** ventral view **E, H** lateral view).

Male genitalia (Fig. 4F–H): Lateral lobes of tegmen moderately elongate, gradually tapering to narrowly rounded apex, which carries long setae. Median lobe stout, median struts more than half length of median lobe; ventral plate longer than dorsal plate; ventral plate with apex widely rounded. Endophallus with tubular structure at basal end.

Female. Size smaller than male, body length 16.67–25.47 mm, humeral width 4.37–7.55 mm. Length (mm) of different body parts: head (3.49) : antenna (32.89)

: pronotum (3.92) : elytra (15.84) : protibia (4.65); width (mm): head (3.20) : pronotum (4.51) : elytra (7.45). Body length/elytral width = 3.13; antenna length/body length = 1.41. Antennomeres with length ratio from base to tip: 6.03 - 1.00 - 14.07 - 9.51 - 8.10 - 7.03 - 6.33 - 5.02 - 4.12 - 3.71 - 5.56.

Habitus is shown in Fig. 1C–D. Eyes (Fig. 3B) with lower lobe shorter than genae. Antennae shortened, extending beyond the elytral apex by five antennomeres. Pronotum (Fig. 3D) slightly wider than long, length/width = 0.87, widest at middle. Variations of pubescence and spots on the elytra is shown in Fig. 2E–H. Protibiae (Fig. 3F) not elongated, without observable tooth at inner side.

Immature stages. Some logs containing larvae were chopped from the type locality and then transferred to the laboratories of Chengdu and Beijing in a constant temperature of 25°C. By observing the pupal chamber (Fig. 5H–K) every day, we observed that *Pseudomacrochenus wusuae* sp. n. took about 28 days from last instar larva (20.I.2016) to pupa (17.II.2016) and about 31 days from pupa to emergence. The habitus of last instar larva is shown in Fig. 5A–D and pupa is shown in Fig. 5E–G.

Host plant. *Craspedolobium schochii* Harms (巴豆藤) (Fig. 6A–B).

Field observations. Biotope in broad-leaved mixed forest of Liangshan Yi Autonomous Prefecture (Sichuan) is shown in Figs 6A–B. Adults in the biotope are shown in Fig. 6C–D.

Remarks. It is easy to distinguish *Pseudomacrochenus wusuae* sp. n. from *P. spinicollis* Breuning, 1949, *P. oberthueri* Breuning, 1955 and *P. albipennis* Chiang, 1981 since the new species has pronotum (Fig. 3C–D) much longer than wide, without long spine at the lateral side, but only with an inconspicuous vestigial small tubercle; while the latter three species have pronotum less elongated, with a distinct spine at each lateral side. In addition, pubescence and spots on the elytra of these species are different.

This new species well resembles *Pseudomacrochenus antennatus* (Gahan, 1894) in general appearance but it is easily distinguishable from it by the combination of the following characters: in *P. wusuae* sp. n., elytra with a large discal spot constituted of pale contrasting grayish setae (Fig. 1E); area around the anterior margin of this spot almost not pubescent, forming a black semicircular ring; tergite VIII (Fig. 4B) with posterior edge weakly emarginate; sternite VIII (Fig. 4B) short and posterior edge more or less truncate; lateral lobes (Fig. 4F) of tegmen with more setae on dorsal surface of apex; ventral plate with apex (Fig. 4G) widely rounded. In *P. antennatus*, elytra without contrasting large spot; tergite VIII (Fig. 4A) with posterior edge distinctly emarginate; sternite VIII (Fig. 4A) longer and posterior edge roundly curved; lateral lobes (Fig. 4C) of tegmen with less setae on dorsal surface of apex; ventral plate with apex (Fig. 4D) rounded.

This new species is also similar to *Pseudomacrochenus affinis* Breuning, 1960, from which it can be distinguished due to the fact that *P. wusuae* sp. n. shows a larger discal spot, with less defined borders, and a hairless black semicircular ring around the ante-

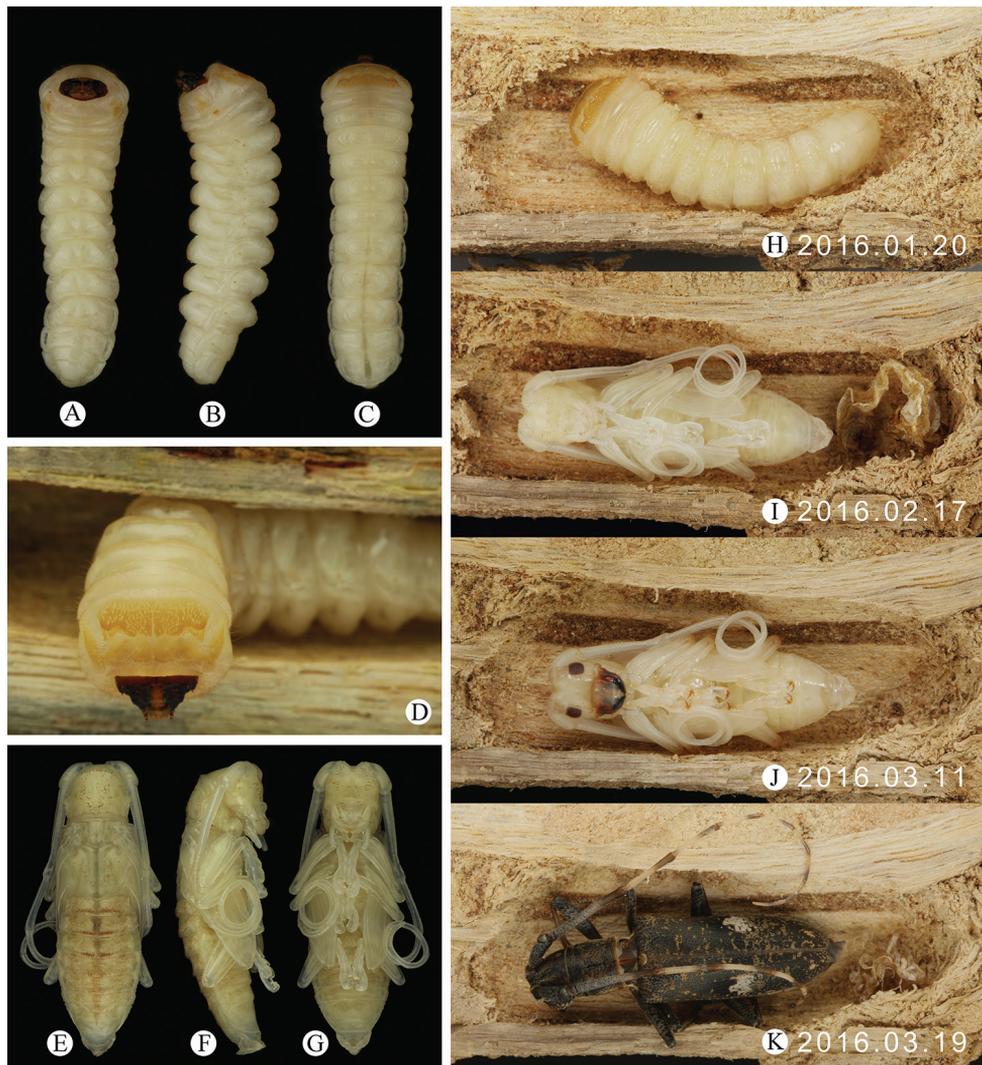


Figure 5. *Pseudomacrochenus wusuae* sp. n. (paratypes). **A–D** last instar larva (**A** ventral view **B** ventro-lateral view **C** dorsal view **D** front view) **E–G** pupa (**E** dorsal view **F** lateral view **G** ventral view) **H** last instar larva in pupal chamber (20.I.2016) **I** pupa in pupal chamber (17.II.2016) **J** pupa in pupal chamber (11.III.2016) **K** newly sclerotized adult in pupal chamber (19.III.2016).

rior margin; while *P. affinis* shows a smaller discal spot, with sharply defined borders, and a quite large hairless black patch before the eytral apex.

Etymology. The specific epithet is dedicated to Ms. Wu-Su Chen, the wife of the first author, for her constant support and love.

Distribution. China (Sichuan).



Figure 6. Field observations of *Pseudomacrochenus wusuae* sp. n. **A** biotope **B** host plant *Craspedolobium schochii* Harms **C** adult (resting) **D** adult (preparing to fly).

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Review of *Dibrachys* Förster from China (Hymenoptera, Chalcidoidea, Pteromalidae)

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Abstract

Twelve species of *Dibrachys* Förster are studied from China, of which four new species, *D. golmudica* Jiao & Xiao, **sp. n.**, *D. kunmingica* Jiao & Xiao, **sp. n.**, *D. liaoi* Jiao & Xiao, **sp. n.** and *D. qinghaiensis* Jiao & Xiao, **sp. n.**, and four newly recorded species, *D. braconidis* (Ferrière & Faure), *D. confusus* (Girault), *D. hians* Bouček and *D. maculipennis* Szélenyi, are reported. A key to Chinese *Dibrachys* and illustrations of external features of the species are provided.

Keywords

China Mainland, *Dibrachys*, key, new species, new record, Pteromalidae, taxonomy

Introduction

Dibrachys was erected by Förster in 1856, but without any species included in the genus. Thomson (1878) subsequently designated *Pteromalus boucheanus* Ratzeburg, 1844 as the type species and listed *Pteromalus cavus* Walker, 1835 as a synonym. Although the type material of *P. boucheanus* is lost (Graham 1969), Thomson's work was accepted by the majority of later researchers. Graham (1969) designated a lectotype for

P. cavus and suggested that, failing the discovery of the type material of *P. boucheanus*, the lectotype of *P. cavus* might conveniently be made also the neotype of *P. boucheanus* because the two were supposed to be identical. However, Peters and Baur (2011) designated a different specimen as lectotype as part of their review of the *Dibrachys cavus* species complex, in which they treated *Dibrachys cavus* as a junior synonym of *Dibrachys microgastri* (Bouché, 1834). Consequently, *D. microgastri* (Bouché) is the senior synonym of both *D. boucheanus* (Ratzeburg) and *D. cavus* (Walker).

Based on differences in mandibular formula and fore wing marginal fringe, Bouček (1965) divided the genus into two subgenera *Dibrachys* Förster s. str. and *D. (Allodibrachys* Bouček). Nineteen valid species of *Dibrachys* are recognized, of which 13 are known from the Palearctic region, 8 from the Nearctic region, 4 from the Oriental region, 1 from the Australasian region, 1 from the Afrotropical region and 2 from the Neotropical region (Noyes 2016). Most species are parasitoids of insect pests, and play an important role in biological control, with 372 different host species being reported for *Dibrachys* (Grissell 1974; Noyes 2016), including species of Lepidoptera, Hymenoptera, Diptera, Coleoptera, Dermaptera, Hemiptera, Neuroptera, Strepsiptera, and several species of Arachnida (Araneidae and Philodromidae). However, as Graham (1969) noted, almost all the host records are associated with *D. cavus*.

Only four species of *Dibrachys* have previously been recorded in China (Liao 1987; Yang 1996). In this study 12 species are identified from China, including four new species (*D. golmudica* Jiao & Xiao, sp. n., *D. kunmingica* Jiao & Xiao, sp. n., *D. liaoi* Jiao & Xiao, sp. n., *D. qinghaiensis* Jiao & Xiao, sp. n.) and four newly recorded species (*D. braconidis* (Ferrière & Faure), *Dibrachys confusus* (Girault), *D. hians* Bouček, and *D. maculipennis* Széleányi).

Material and methods

A total of 943 specimens was examined from the museum of Institute of Zoology, the Chinese Academy of Sciences (IZCAS). All material from our own collection was swept and preserved in 75% ethanol. Specimens were subsequently air dried, point-mounted and examined with a Nikon SMZ1500 stereomicroscope. Photographs were taken using a Nikon Multizoom AZ100 system, and the plates were compiled using Adobe Photoshop CS3 software. In addition, the author had examined specimens of *Dibrachys* deposited in the National History Museum, London, the Naturalis, Leiden and the Bavarian State Collections of Zoology in April, 2002.

Morphological terminology mostly follows that of Graham (1969), Bouček (1988) and Gibson (1997). All specimens were examined and identified using the keys of Graham (1969), Grissell (1974), Doganlar (1987), Yang (1996), Zerova et al (1992) and Peters and Baur (2011). Every new species is described based on the holotype specimen, other species are described basing on the examined material available to us. Body length excludes the ovipositor sheaths and is measured in millimeters (mm); other measurements are given as ratios.

Abbreviations of morphological terms used are:

- Fu_n** funicular segment number;
- POL** posterior ocellar distance;
- OOL** ocellocular distance;
- Gt_n** gastral tergum number.

Taxonomy

Key to species

- 1 Left mandible with three teeth and right mandible with four teeth (Fig. 6); fore wing with marginal fringe except between marginal vein and wing apex; occipital carina transverse, closer to foramen than vertex; gaster mostly ovate (Figs 1, 5), slightly longer than broad ***D. (Allodibrachys)*...2**
- Both mandibles with four teeth (Fig. 21); fore wing without marginal fringe; occipital carina curving, closer to vertex than foramen; gaster spindle-shaped (Figs 15, 23, 33, 38), distinctly longer than broad ***D. (Dibrachys)*...6**
- 2 Head in frontal view with gena almost straight and with lower angle of gena protruding beyond clypeal margin (Fig. 3)..... ***D. hians***
- Head in frontal view with gena evenly curved and with lower angle of gena not exceeding clypeal margin (Figs 6, 12) **3**
- 3 Stigmal vein longer than postmarginal vein; gaster 1.5 times as long as broad ***D. koraiensis***
- Stigmal vein shorter than or at most as long as postmarginal vein; gaster at most 1.3 times as long as broad..... **4**
- 4 Lower margin of clypeus not protruded, slightly emarginate in middle and without tooth (Fig. 6); head in dorsal view with POL 1.33 times as long as OOL ***D. kojimae***
- Lower margin slightly protruded, emarginate in middle and with two obtuse teeth; head in dorsal view with POL more than 1.5 times as long as OOL **5**
- 5 Marginal vein 1.5 times as long as stigmal vein; propodeum with plica complete and median carina only distinct basally ***D. yunnanensis***
- Marginal vein 1.91 times as long as stigmal vein; propodeum with plica only conspicuous basally, median carina absent ***D. kunmingica* sp. n.**
- 6 Antennae with three anelli; postmarginal vein very shorter than stigmal vein, at most half length of stigmal vein ***D. golmudica* sp. n.**
- Antennae with two anelli; postmarginal vein longer or slightly shorter than stigmal vein **7**
- 7 Lower margin of clypeus broadly emarginate, without tooth; gaster 1.37 times as long as broad ***D. braconidis***
- Lower margin of clypeus slightly protruding, emarginate in middle and with two blunt or sharp teeth; gaster at least 1.8 times as long as broad **8**

- 8 Antennal insertion slightly above lower ocular line, antennal scape reaching lower margin of anterior ocellus..... **9**
- Antennal insertion place on lower ocular line, antennal scape not reaching lower margin of anterior ocellus..... **11**
- 9 Fore wing with a yellowish-brown infumation behind marginal vein; propodeum with incomplete median carina; stigmal vein slightly longer than postmarginal vein; gaster slightly broader than thorax width ***D. maculipennis***
- Fore wing immaculate, without any infumation; propodeum with complete median carina; stigmal vein as long as postmarginal vein; gaster narrower than thorax width **10**
- 10 Antennal scape as long as eye height; head in frontal view 1.27 times as wide as high ***D. qinghaiensis* sp. n.**
- Antennal scape distinctly shorter than eye height; head in frontal view 1.15 times as wide as high..... ***D. confusus***
- 11 Lower margin of clypeus emarginate in middle and with two sharp teeth (Fig. 31); antennae with Fu_1 to Fu_4 slightly longer than its broad; head in dorsal view 1.8 times as wide as long; gaster 1.8 times as long as broad.....
- ***D. liaoi* sp. n.**
- Lower margin of clypeus emarginate in middle and with two blunt teeth (Figs 40, 45); antennae with Fu_1 to Fu_4 quadrate; head in dorsal view 2 times as wide as long; gaster 2 times as long as broad ***D. microgastri***

***Dibrachys* Förster, 1856**

Dibrachys Förster, 1856: 65. Type-species: *Pteromalus boucheanus* Ratzeburg, designated by Thomson 1878: 47 (= *Diplolepis microgastri* Bouché, 1834: 168).

Dibrachys Förster: Dalla Torre 1898: 155; Graham 1969: 804–814; Wallace 1973: 175–176; Bouček 1993: 1259; Yang 1996: 196–201, 323–324.

Coelopisthoidea Gahan, 1913: 178–183. Type-species: *Coelopisthoidea cladiae* Gahan, 1913: 178–183. Synonymized by Girault 1916b: 408; Bouček 1988: 434.

Diagnosis. Body dark green. Head in frontal view round; antennal insertion placed on lower ocular line and face not protuberant at antennal insertion; antennal formula 11263 (rarely 11353); lower margin of clypeus with two sinuate teeth; both mandibles with four teeth or right mandible with four teeth and left mandible with three teeth; head in dorsal view with occiput margined by blunt or sharp, transverse ridge. Mesosoma slightly convex; pronotal collar not margined or slightly carinate medially; notauli incomplete and inconspicuous; scutellum without frenal groove; propodeum with plica complete, median carina developed or not. Fore wing without marginal fringe or at least bare between postmarginal vein and wing apex; postmarginal vein short, only inconspicuously longer than stigmal vein. Hind tibia with one spur. Gaster ovate.

Distribution. Widespread world-wide distribution, see Noyes (2016). China: Heilongjiang, Jilin, Liaoning, Inner Mongolia, Beijing, Hebei, Shanxi, Shandong, Henan, Shaanxi, Ningxia, Gansu, Qinghai, Xinjiang, Jiangsu, Shanghai, Anhui, Zhejiang, Hubei, Jiangxi, Hunan, Guangxi, Sichuan, Guizhou, Yunnan and Tibet.

Descriptions of species

Dibrachys (*Allodibrachys*) **Bouček**

Dibrachys sgen. *Allodibrachys* Bouček, 1965: 30. Type-species : *Dibrachys hians* Bouček, by original designation.

Diagnosis. The subgenus have the left mandible with three teeth and right mandible with four teeth; occipital carina transverse, closer to foramen than vertex; fore wing with marginal fringe except between marginal vein and wing apex; gaster mostly ovate, slightly longer than broad.

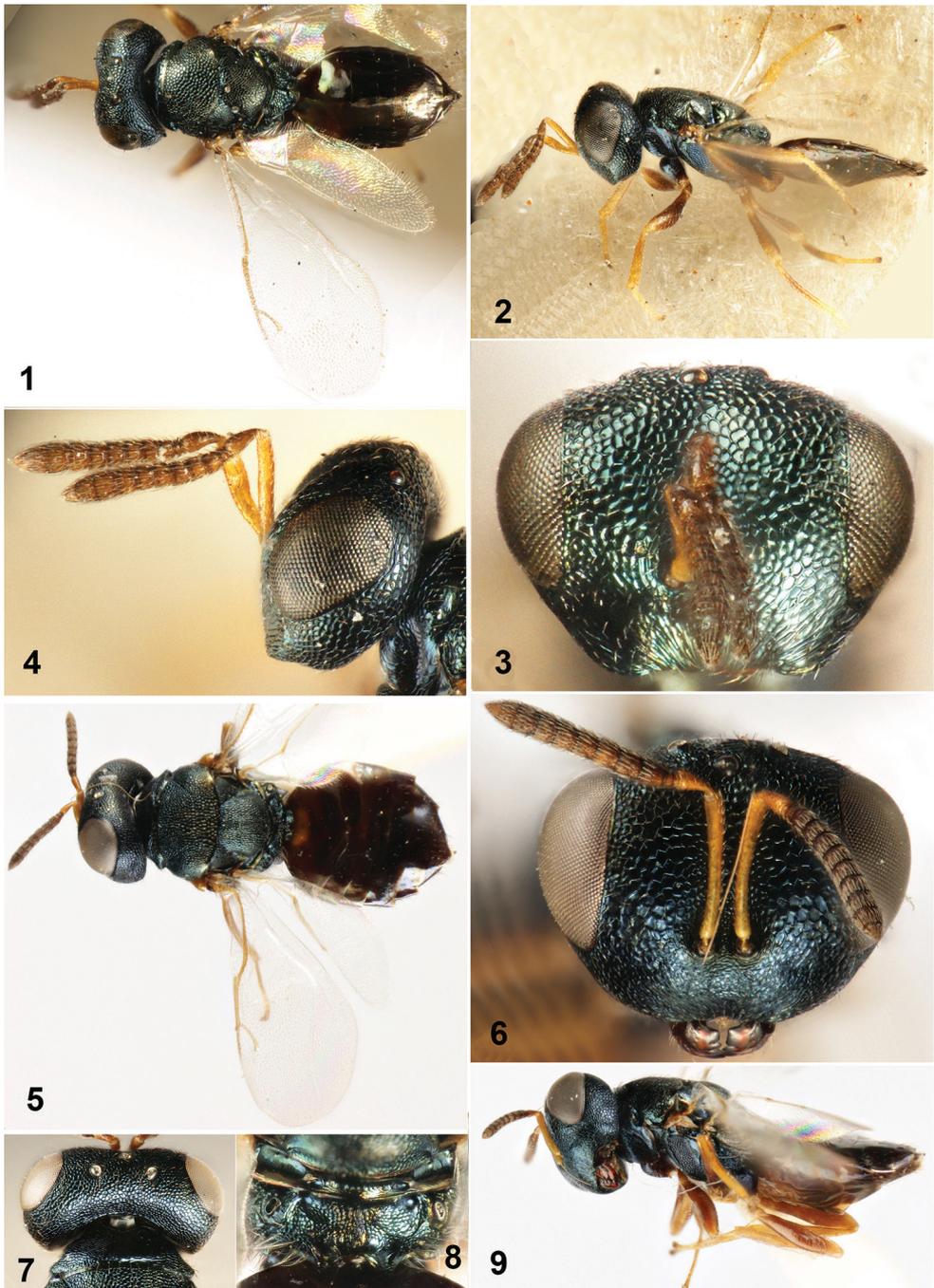
Dibrachys hians Bouček, 1965, new record to China

Figs 1–4

Dibrachys (*Allodibrachys*) *hians* Bouček, 1965: 28.

Diagnosis. Body length 1.6–2.0 mm (Figs 1, 2). Head in frontal view (Fig. 3) 1.32 × as wide as high; antennal scrobe shallow, extending upwards and not reaching anterior ocellus; antennal insertion placed on lower ocular line; clypeus with longitudinal striation; lower margin of clypeus not protruded, emarginate in middle, and without tooth; gena almost straight, lower angle of gena protruding beyond clypeal margin. Antenna (Fig. 4) with scape shorter than eye height (0.79×), not reaching anterior ocellus; length of pedicel and flagellum combined less than head width (0.67×); anelli transverse; each funicular segment subquarate; clava slightly clavate. Head in dorsal view with width 2× length; eye length 2× temple length; POL 2× OOL. Mesosoma 1.37× as long as broad, mid lobe of mesoscutum with regular sculpture. Propodeum with complete median carina and incomplete plica. Fore wing with marginal vein 1.8× as long as postmarginal vein; postmarginal vein as long as stigmal vein. Gaster 1.25× as long as broad, slightly broader than thorax width.

Material examined. China: 3♀, Heilongjiang: Yichun, 13.VII.1962, ex. Tachinidae sp. on *Ptycholomoides aeriferanus* Herrich-Schaffer, leg. Ding-Xi Liao; 1♂, 1♀, Heilongjiang: Dailing, 29.VI.1962, ex. Tachinidae sp. on *Ptycholomoides aeriferanus* Herrich-Schaffer; 1♀, Jilin: Dunhua, 25.VI.1990; 1♂, 2♀, Beijing: Yanqing, 19.VII.1982, ex. Curculionidae sp. on elm, leg. Ding-Xi Liao.



Figures 1–9. 1–4 *Dibrachys hians* Bouček, 1965. 1 Body in dorsal view 2 Body in lateral view 3 Head in frontal view 4 Head in lateral view 5–9 *Dibrachys kojimae* (Ishii, 1916) 5 Body in dorsal view 6 Head in frontal view 7 Head in dorsal view 8 Propodeum 9 Body in lateral view.

Hosts. Trematerra (1988) reported *Pyralis farinalis* L (Lepidoptera: Pyralidae). as a host and here we record Curculionidae sp. on elm, Tachinidae sp. on *Ptycholomoides aeriferanus* Herrich-Schaffer (Lepidoptera: Tortricidae), and Tachinidae sp. on *Laspeyresia grunertiana* (Ratzeburg) (Lepidoptera: Noctuoidea).

Distribution. China (Heilongjiang, Jilin, Beijing); Palearctic and Nearctic regions.

***Dibrachys kojimae* (Ishii, 1938)**

Figs 5–9

Euterus kojimae Ishii, 1938: 100.

Dibrachys kojimae (Ishii): Kamijo 1982: 74.

Diagnosis. Body length 2.5–3.2 mm (Figs 5, 9). Head in frontal view (Fig. 6) 1.31× as wide as high; sculpture on lateral area of antennal scrobe distinctly larger than on vertex and face; lower margin of clypeus not protruded, slightly emarginate in median; gena evenly curved, lower angle of gena not exceeding clypeal margin. Antenna with scape shorter than eye height (0.81×); length of pedicel and flagellum combined less than head width (0.94×); anelli transverse and second anellus longer than first anellus; Fu_1 - Fu_3 slightly long than wide, Fu_4 - Fu_6 subquadrate. Head in dorsal view (Fig. 7) 2× as wide as long; eye length 1.67× temple length, POL 1.33× OOL. Mesosoma 1.32× as long as broad; with regular sculpture. Propodeum (Fig. 8) with incomplete median carina and complete plica. Fore wing with submarginal vein 2.3× as long as marginal vein; marginal vein 1.6× as long as postmarginal vein, 1.8× as long as stigma vein; stigmal vein slightly shorter than postmarginal vein (0.9×). Gaster 1.2× as long as broad, slightly broader than thorax width.

Material examined. China: 2♂, 16♀, Beijing: Miyun Reservoir, 10-20.VII.1983, ex. pupae of *Dendrolimus tabulaeformis* Tsai et Liu, leg. Ju-Wen Wu; 7♀, Henan: Fangchen, Dalai, VIII. 1983, ex. pupae of *Dendrolimus tabulaeformis* Tsai et Liu, leg. De-Long Shui; 1♂, 11♀, Anhui: Dongzhi, 1983, ex. pupae of *Dendrolimus*, leg. Ding-Xi Liao; 2♂, 3♀, Anhui: Dongzhi, 7.V.1983, ex. *Dendrolimus punctatus* Walker, leg. Ding-Xi Liao; 3♀, Anhui: Qianshan, Tianzhu Mountain, 4.IX.1976, ex. pupae of *Dendrolimus*, leg. Tao-Qian Hou; 1♀, Hubei: Wuhan, 25.VI.1980, ex. *Dendrolimus punctatus* (Walker), leg. Tao-Qian Hou; 4♀, Hunan, 25.X.1979, ex. pupae of *Pieris brassicae* L., leg. Ding-Xi Liao; 1♂, 7♀, Hunan: Chengbu, 16. VIII.1986, ex. pupae of *Dendrolimus kikuchii* Matsumura, leg. Zheng-Mao Li; 10♂, 15♀, Hunan: Daoxian, 29.XII.1979, ex. eggs of *Lebeda nobilis* Walker, leg. Ding-Xi Liao; 1♂, 5♀, Hunan: Daoxian, 12.XI.1973, ex. eggs of *Lebeda nobilis* Walker, leg. Xin-Wang Tong; 2♀, Guangxi: Nanning, VI.1975, ex. Eggs of *Dendrolimus*, leg. Lin Wei; 2♀, Guizhou: Anshun, 25.X.1980, ex. pupae of *Dendrolimus houi* Lajonquiere, leg. Jin-Rong Zhou; 6♀, Yunnan: Baoshan, 21.v.1975, ex. *Dendrolimus*, leg. Ding-xi Liao; 1♀, Tibet: Mêdog, 1100m, 26.I.1983, leg. Yin-Heng Han.

Hosts. Kamijo (1982) reported *Dendrolimus spectabilis* Butler (Lepidoptera: Lasiocampidae) as a host and here we report *Dendrolimus tabulaeformis* Tsai et Liu, pupae/eggs of *Dendrolimus*, *Dendrolimus punctatus* (Walker), *Dendrolimus kikuchii* Matsumura, *Dendrolimus houi* Lajonquiere, *Lebeda nobilis* Walker (Lepidoptera: Lasiocampidae) and *Pieris brassicae* L. (Lepidoptera: Pieridae).

Distribution. China (Beijing, Anhui, Jiangxi, Henan, Hunan, Guangxi, Guizhou, Yunnan, Tibet); Japan.

Dibrachys koraiensis Yang, 1996

Dibrachys koraiensis Yang, 1996: 197–199, 323.

Diagnosis. Body length 2.5–2.7 mm; gaster long ovate. Head in frontal view 1.25× as wide as high; antennal scrobe extending upwards and reaching anterior ocellus; lower face slightly convex; antennal insertion placed on lower ocular line; clypeus with longitudinal striation and lower margin slightly protruded, emarginate, and with two blunt teeth; lower angle of gena not exceeding clypeal margin. Antennal scape slightly shorter than eye height (0.87×); length of pedicel and flagellum shorter than head width (0.8×); anelli transverse; Fu_1 and Fu_2 slightly longer than broad, Fu_3 and Fu_4 quadrate, Fu_4 and Fu_6 slightly transverse. Head in dorsal view 1.88× as wide as long; eye length 2× temple length; POL 1.5× OOL. Mesosoma 1.7× as long as broad, mid lobe of mesoscutum with relatively coarse sculpture. Propodeum with complete plica and indistinct median carina. Fore wing length 2.2× width; submarginal vein 2× as long as marginal vein; marginal vein 2.53× as long as postmarginal vein, 2.13× as long as stigma vein; stigmal vein slightly longer than postmarginal vein (1.15×). Gaster 1.5× as long as broad, distinctly broader than thorax width (1.21×).

Material examined. China: 1♀, Heilongjiang: Yichun, 3.VII.1972, leg. Ding-xi Liao; 1♀, Heilongjiang: Hailin, VI.1975, leg. Gui-you Zhang.

Hosts. Yang (1996) reported this species as reared from the pupae of some chalcid collected from tunnels in *Picea koraiensis* Nakai (Pinales: Pinaceae) built by the wood pest *Orthotomicus golovjankoi* Pjatnitzky (Coleoptera: Curculionidae), the possible host.

Distribution. China (Heilongjiang).

Dibrachys kunmingica Jiao & Xiao, sp. n.

<http://zoobank.org/DC6570D7-7010-4813-89D4-570403E3A730>

Figs 10–14

Diagnosis. The species belongs to subgenus *Allodibrachys*, and similar to *D. yunnanensis* Yang has the lower angle of the gena not exceeding the clypeal margin, and the stigmal vein slightly shorter than the postmarginal vein. The main differences are:

marginal vein $2\times$ as long as stigmal vein; propodeum with plica indistinct, only conspicuous basally; median carina absent.

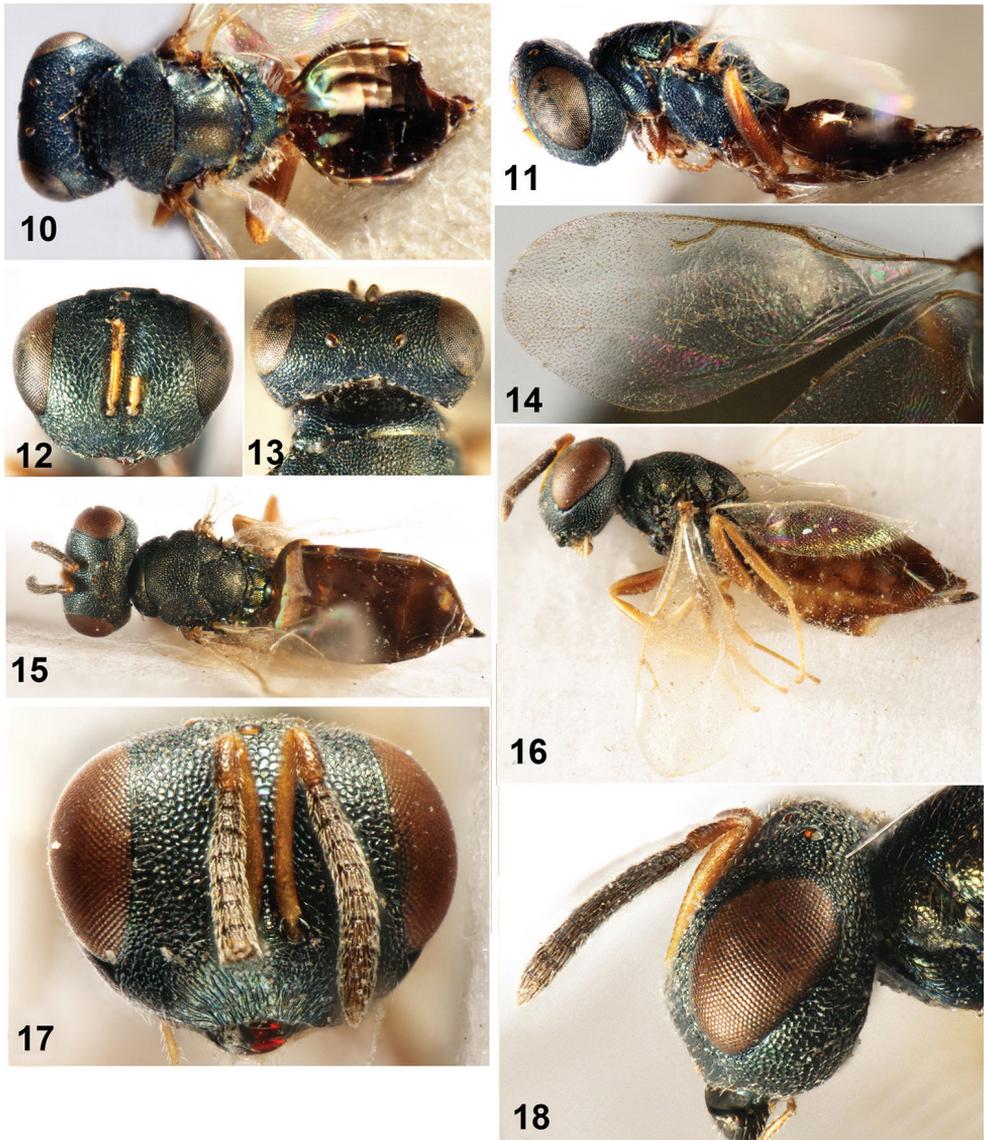
Description. Holotype. *Female.* Body (Figs 10, 11) length 2.5 mm. Head and mesosoma dark green, with metallic reflection; gaster brown with metallic reflection basally. Antenna brown except pedicel and scape yellowish brown; legs light brown except coxae brown; fore wing hyaline, slightly infumate, wing venation yellowish-brown.

Head in frontal view (Fig. 12), width $1.29\times$ height; frons with irregular reticulation, lower face curved ventrally; eye height $0.68\times$ head height, inner margin of eyes slightly converging upwards, separated by $1.24\times$ their height; antennal scrope deep, not reaching anterior ocellus; reticulation in antennal scrope smaller than that on parascrope. Antennal insertion slightly above lower ocular line, distance from upper margin of antennal torulus to lower margin of anterior ocellus $2\times$ distance from lower margin of antennal torulus to clypeal margin; clypeus with dense longitudinal striation; clypeal margin slightly protruded, emarginate in the middle and with two blunt teeth, median margin concave, as a small, smooth, triangular depression; gena plump, oral fossa $0.48\times$ as wide as head. Head in lateral view with malar sulcus inconspicuous, eye height $2.2\times$ malar space. Antennal scape length $0.83\times$ eye height, not reaching anterior ocellus; pedicel in lateral view $2.5\times$ as long as broad; both anelli transverse. Head in dorsal view (Fig. 13), head $2\times$ as wide as long; vertex convex, with regular reticulation denser than that on frons, posterior part sharply sloped down; eye length $2\times$ temple length; POL $1.6\times$ OOL.

Head $1.19\times$ as broad as thorax. Mesosoma $1.37\times$ as long as broad. Propodeum with short collar, collar subhorizontal and not margined, posterior margin smooth. Mesoscutum $2\times$ as broad as long, reticulation on posterior area bigger than that on anterior area. Scutellum slightly convex medially, width $1.22\times$ length, frenal line absent; reticulation shallower than that on mesoscutum posteriorly. Median length of propodeum half that of scutellum; median area flat, with deep, fine, dense reticulation; median carina absent; plica incomplete, visible anteriorly; plicae separated by $1.68\times$ median length of propodeum; short nucha hemispheric and smooth; spiracles elongate, $2\times$ as long as broad, separated from hind margin of metanotum by width of spiracle. Fore wing (Fig. 14) $2.36\times$ as long as broad; without fringe from postmarginal vein to distal margin; hind wing with marginal fringe; basal vein and basal cell bare, speculum only extending to base of marginal vein; upper surface of costal cell bare, lower surface with one compact row of setae and distal $1/3$ with one row of short setae and some scattered setae; submarginal vein $2\times$ as long as marginal vein; marginal vein $1.91\times$ as long as postmarginal vein, $2\times$ as long as stigma vein; stigmal vein slightly shorter than postmarginal vein ($0.96\times$); stigmal vein curved.

Petiole quadrate, as long as broad. Gaster (Fig. 10) ovate, $1.2\times$ as long as broad, width $1.06\times$ thorax width, length $0.94\times$ mesosoma length; Gt_1 covering $0.42\times$ length of gaster, posterior margin of Gt_1 cambered, without distinct fovea in the middle; following tergites with posterior margin straight; tergites coriaceous.

Material examined. Holotype. ♀, China: Yunnan: Kunming, 25.94°N , 102.42°E , IV.1954, leg. Ding-Xi Liao. Paratype. 1 ♀, same data as holotype.



Figures 10–18. 10–14 *Dibrachys kunmingica* sp. n., female holotype **10** Body in dorsal view **11** Body in lateral view **12** Head in frontal view **13** Head in dorsal view **14** Fore wing **15–18** *Dibrachys bracidis* (Ferrière et Faure, 1925). **15** Body in dorsal view **16** Body in lateral view **17** Head in frontal view **18** Head in lateral view.

Etymology. Named after the location of the type material.

Hosts. Unknown.

Distribution. China (Yunnan).

***Dibrachys yunnanensis* Yang, 1996**

Dibrachys yunnanensis Yang, 1996: 199–201, 324.

Diagnosis. Body squat, length 2.0–2.4 mm. Head in front view 1.3× as wide as high; antennal scrobe shallow, extending upwards and reaching anterior ocellus; antennal insertion placed on the lower ocular line; clypeus with longitudinal sculpture; lower margin of clypeus slightly protruded, median emarginate and with 2 sinuate teeth; gena evenly curved, lower angle of gena not exceeding clypeal margin. Antennal scape slightly shorter than eye height, length of flagellum and pedicel combined less than head width (0.74×); pedicel in lateral view 2.2× as long as broad; both anelli transverse; Fu_1 and Fu_5 quadrate, Fu_6 distinctly transverse. Head in dorsal view, 1.9× as wide as long, eye length 1.79× temple length; POL 1.78× OOL. Mesosoma 1.5× as long as broad, mid lobe of mesoscutum with regular sculpture. Propodeum with plicae complete, median carina distinct on base part. Fore wing with submarginal vein more than 2× as long as marginal vein, marginal vein 1.5× as long as postmarginal vein, stigmal vein as long as postmarginal vein. Gaster 1.3× as long as broad, as broad as thorax width.

Material examined. China: 1♀, Yunnan: Nanjian, 2.VI.1980, leg. Ding-Xi Liao.

Hosts. This species parasitized on larvae and pupae of *Tomicus piniperda* L. (Coleoptera: Curculionidae) which harmful to *Pinus yunnanensis* (Pinales: Pinaceae) (Yang 1996).

Distribution. China (Yunnan).

Dibrachys (Dibrachys) Förster

Diagnosis. Both mandibles with four teeth; occipital carina curving, closer to vertex than foramen; fore wing without marginal fringe; gaster spindle-shaped, distinctly longer than broad.

***Dibrachys braconidis* (Ferrière & Faure, 1925), new record to China**

Figs 15–18

Homoporus luniger braconidis Ferrière & Faure, 1925, 11: 226.

Dibrachys braconidis (Ferrière & Faure): Bouček 1965: 30; Viggiani 1968: 112–115.

Diagnosis. Body slender, length 2.0–2.9 mm (Figs 15, 16). Head in frontal view, 1.3× as wide as high; antennal scrobe extending to anterior ocellus; antennal insertion slightly above lower ocular line; lower face at least slightly convex; clypeus with longitudinal striation, lower margin of clypeus not protruding (Figs 17, 18), without tooth. Antennal scape slightly shorter than eye height, length of pedicel and flagellum shorter than head width (0.8×); Fu_1 to Fu_3 slightly longer than broad, Fu_4 to Fu_6

quadrate. Head in dorsal view, 1.9× as wide as long; eye length 2× temple length; POL 1.44× OOL. Mesosoma 1.37× as long as broad. Propodeum with complete median carina and plicae. Fore wing 2.33× as long as broad; submarginal vein 2.24× as long as marginal vein; marginal vein 1.9× as long as postmarginal vein; stigmal vein as long as postmarginal vein. Gaster 1.37× as long as broad, 1.27× as broad as thorax width.

Material examined. China: 30♂, 29♀, Sichuan: Xichang, V.1992, ex. *Neodiprion xiangyunicus* Xiao et Zhou, leg. Zhen Zhang; 1♀, Yunnan: Nanjian, 2.VI.1980; Yunnan: Kunming, 13.VII.1977, ex. pupae of Diprioninae, leg. Jing-liang Qi; 2♀, Yunnan: Kunming, XII.1988, leg. Hong-ming Yang; 1♀, Tibet: Chamdo, 3400m, 15.VIII.2001, leg. Chao-dong Zhu.

Hosts. The species mainly parasitizes *Luffia ferchaultella*, and *Luffia lapidella* (Lepidoptera: Psychidae) and *Apanteles glomeratus* (L.) (Hymenoptera: Braconidae) (Graham, 1969). Here we newly report *Neodiprion xiangyunicus* Xiao et Zhou and Diprionidae sp. (Hymenoptera: Symphyta).

Distribution. China (Sichuan, Yunnan, Tibet); Palearctic and Nearctic regions. This is the first record from the Oriental region.

***Dibrachys confusus* (Girault, 1916), new record to China**

Figs 19–22

Coelopisthia confusus Girault, 1916a: 246.

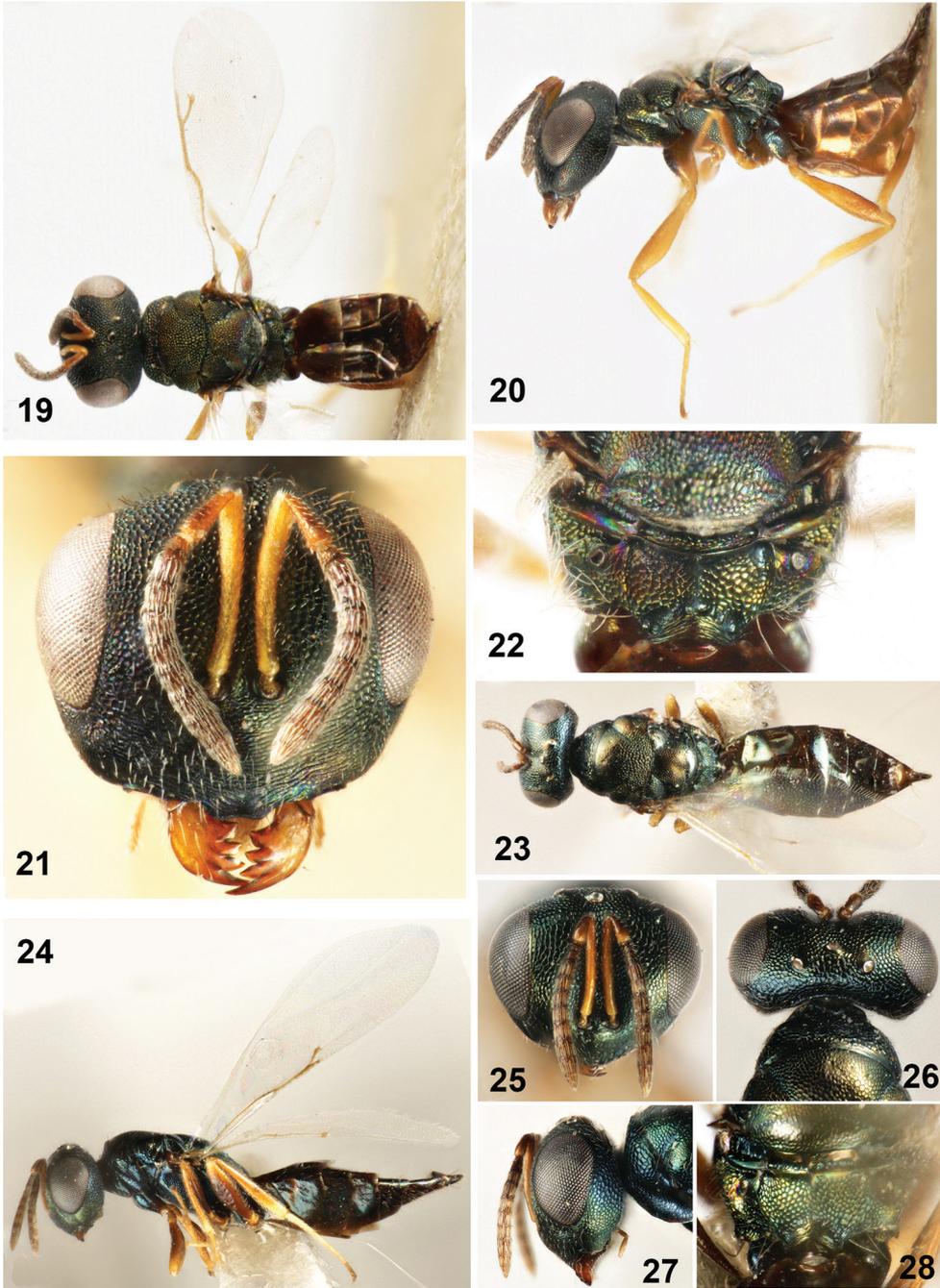
Dibrachys confusus (Girault): Peck 1951: 554; Grissell 1974: 318; Burks 1979: 828.

Diagnosis. Body slightly slender (Figs 19, 20), about 2.6 mm. Head in frontal view (Fig. 21) 1.15× as wide as high; antennal scrobe extending upwards and not reaching anterior ocellus; antennal insertion slightly above lower ocular line; lower face flat; clypeus with transverse striation and lower margin protruding, emarginate with two blunt teeth. Antennal scape slightly shorter than eye height (0.91×) but reaching lower margin of anterior ocellus; length of pedicel and flagellum combined shorter than head width; anelli transverse; each funicular segment slightly longer than its broad respectively. Head in dorsal view with width 2× length; eye length 1.87× temple length; POL 1.46× OOL. Mesosoma 1.43× as long as broad, mesoscutum with regular sculpture. Propodeum with median carina complete (Fig. 22), plicae complete and parallel anteriorly. Fore wing 2.38× as long as broad; submarginal vein 2.37× as long as marginal vein; marginal vein 1.72× as long as postmarginal vein; stigmal vein as long as postmarginal vein. Gaster 2× as long as broad, narrower than thorax width.

Material examined. China: 3♀, Beijing: Yuanmingyuan Imperial Garden, 2.VI.1984, ex. larvae of *Lymantria dispar* (L.), leg. Ding-Xi Liao.

Hosts. De Santis (1983) reported *Megachile rotundata* (Fabricius) (Hymenoptera: Megachilidae) as a host and here we record the larvae of *Lymantria dispar* (L.) (Lepidoptera: Lymantriidae).

Distribution. China (Beijing); Palearctic, Nearctic and Neotropical regions.



Figures 19–28. 19–22 *Dibrachys confusus* (Girault, 1916) 19 Body in dorsal view 20 Body in lateral view 21 Head in frontal view 22 Propodeum 23–28 *Dibrachys golmudica* sp. n., female holotype 23 Body in dorsal view 24 Body in lateral view 25 Head in frontal view 26 Head in dorsal view 27 Head in lateral view 28 Propodeum.

***Dibrachys golmudica* Jiao & Xiao, sp. n.**

<http://zoobank.org/F39A82B6-44C1-4E9A-9D23-B4462E870754>

Figs 23–28

Diagnosis. The new species belongs to *Dibrachys s. str.*, and the noticeable differences with other species of the subgenus by the following characters: in female, antennae with three anelli; the postmarginal vein being distinctly shorter than the stigmal vein ($0.5\times$), and gaster being $2.5\times$ as long as broad.

Description. Holotype. *Female.* Body length 2.2 mm (Figs 23, 24). Head and mesosoma black with bronze luster and metallic reflection. Gaster dark brown with metallic reflection basally. Antennal scape yellowish brown to light brown from base to apex, flagellum dark brown; legs yellowish brown except coxae concolorous with body and femora light brown; fore wing hyaline, without infumation, venation yellowish brown or yellowish.

Head in frontal view (Fig. 25) $1.24\times$ as wide as high; eyes with inner margins parallel, eye height $0.62\times$ head height, eyes separated by $1.26\times$ their height; lower face with weak striation, upper face with obvious regular reticulation; antennal scrobe deep, not reaching anterior ocellus. Antennal insertion on lower ocular line, distance from upper margin of torulus to lower margin of anterior ocellus $2.54\times$ distance from lower margin of torulus to lower margin of clypeus; clypeus with longitudinal striation on both sides, lower margin slightly protruded, emarginate in middle with two obtuse teeth; oral fossa $0.49\times$ as wide as head. Head in lateral view (Fig. 27) with malar sulcus conspicuous, eye height $1.5\times$ its broad and $2.83\times$ malar space. Antennal scape length $0.91\times$ eye height; length of flagellum and pedicel combined less than head width ($0.88\times$); pedicel in lateral view $2.3\times$ as long as broad; antenna with 3 anelli, Fu_1 and Fu_2 distinctly transverse, Fu_3 quadrate, Fu_1 to Fu_3 combined $0.78\times$ as long as pedicel; Fu_4 longer than broad, Fu_5 quadrate; each funicular segment with one row of sensilla; setae on funicle all decumbent; clava not distinctly clavate, $3.4\times$ as long as broad, micropilosity limited to apex of third clava segment. Head in dorsal view (Fig. 26), $2\times$ as wide as long; vertex convex, sculpture on vertex slightly smaller than sculpture on frons; occipital carina distinct; eye length $2.5\times$ temple length; POL $2.11\times$ OOL.

Head $1.24\times$ as broad as thorax. Mesosoma $1.6\times$ as long as broad. Pronotum $0.65\times$ as broad as mesoscutum, collar rounded, posterior band smooth. Mesoscutum $1.57\times$ as broad as long, with regular reticulation, in anterior half weakly reticulate and posterior half with deep reticulation; notauli distinct but not complete. Scutellum convex, $1.07\times$ as broad as long, frenal line absent; reticulation smaller than on mesoscutum, but regular and shallow. Propodeum medially $\frac{1}{2}$ as long as scutellum, with fine, deep, dense reticulation; plica weak (Fig. 28), only visible basally and separated by $2\times$ medial length of propodeum; median carina incomplete; propodeum with short, slightly convex nucha having transverse striation; propodeal spiracles elongate, $2.67\times$ as long as broad. Fore wing $2.16\times$ as long as broad, without marginal fringe; setae pale, inconspicuous; basal vein and basal cell bare, upper surface of costal cell bare, lower surface with one complete row of setae and distally with some scattered setae; submarginal

vein $2.75\times$ as long as marginal vein, marginal vein $2.63\times$ as long as stigmal vein, post-marginal vein shorter than stigmal vein (at most $0.5\times$); stigmal vein slightly curved.

Gaster spindle-shaped with apex pointed (Fig. 23), $2.5\times$ as long as broad; as wide as thorax; Gt_1 covering $1/4$ of gaster, with posterior margin cambered; tergites beyond Gt_1 equal in length; ovipositor exerted.

Male. Head black except frons with yellowish-green, and antennae yellow; mesosoma black except thorax purplish laterally, legs yellow except coxae brown. Antennae with two distinctly transverse anelli, pedicel in lateral view $1.8\times$ as long as broad, each funicular segment longer than broad; gaster oval, apex not pointed.

Material examined. Holotype. ♀, China: Qinghai: Golmud, Guolemude, 2880m, 36.26°N , 94.53°E , 14.IX.2001, leg. Chao-Dong Zhu. Paratype. 1♀, same data to holotype; 3♂, 6♀, Inner Mongolia: Ejin B., 11.VI.1981, ex. *Dinorhopala* on *Populus diversifolia*, leg. Hua-Qiang Shao.

Etymology. Named after the location where the holotype was collection.

Hosts. Specimens from Inner Mongolia were reared from *Dinorhopala* (Coleoptera: Curculionidae) on *Populus diversifolia*.

Distribution. China (Inner Mongolia, Qinghai).

Dibrachys liaoi Jiao & Xiao, sp. n.

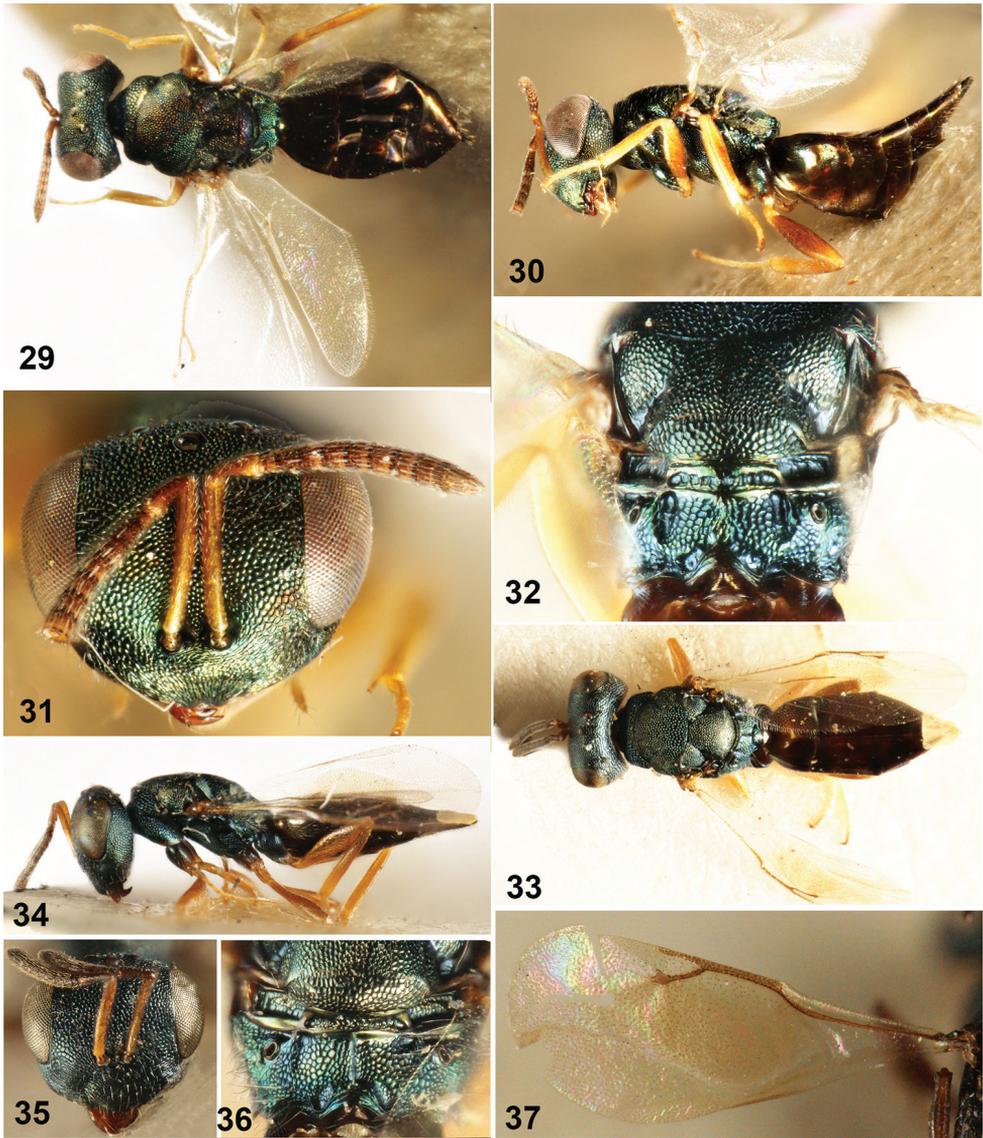
<http://zoobank.org/2418A0BD-799F-44D4-8A3B-BCCAB3FCF7C2>

Figs 29–32

Diagnosis. The new species belongs to *Dibrachys* s. str., and the mainly differences with *Dibrachys microgastri* (Bouché) as follows: *D. liaoi* sp. n. slightly blue-greenish, clypeal margin with two sharp teeth, Fu_1 to Fu_4 length slight longer than its width respectively, Fu_5 and Fu_6 quadrate, gaster $1.8\times$ as long as broad; but in *D. microgastri* (Bouché), body yellow-green, clypeal margin with two blunt teeth, Fu_1 to Fu_5 quadrate, Fu_6 transverse, gaster $2\times$ as long as broad.

Description. Holotype. *Female.* Body (Figs 29, 30) length 2.2 mm. Head and mesosoma dark green, with metallic reflection; gaster brown and with metallic reflection basally. Antennae dark brown except scape and pedicel yellowish brown; mandible brown; legs yellowish brown except coxae brown; fore wing slightly infumate, wing venation yellowish brown.

Head in frontal view (Fig. 31), width $1.24\times$ height; frons with dense reticulation; lower face flat, reticulation on lower face same as that on frons; eye height $0.7\times$ head height, eyes separated by $1.09\times$ eye height; antennal scrobe deep, extending upwards but not reaching anterior ocellus. Antennal insertion on lower ocular line, distance from upper margin of torulus to lower margin of anterior ocellus $2.35\times$ distance from lower margin of torulus to clypeal margin; clypeus with longitudinal sculpture, only small area smooth; clypeal margin protruded, emarginate in middle with two sharp teeth; oral fossa width $0.46\times$ head width. Head in lateral view, malar sulcus inconspicuous, eye height $3.3\times$ malar space. Antennal scape $0.81\times$ as long as eye height, not reaching lower margin



Figures 29–37. 29–32 *Dibrachys liaoi* sp. n., female holotype 29 Body in dorsal view 30 Body in lateral view 31 Head in frontal view 32 Propodeum 33–37 *Dibrachys maculipennis* Szelenyi 33 Body in dorsal view 34 Body in lateral view 35 Head in frontal view 36 Propodeum 37 Fore wing.

of anterior ocellus; length of pedicel and flagellum combined shorter than head width ($0.84\times$); pedicel in lateral view $2.6\times$ as long as broad; anelli transverse; Fu_1 to Fu_4 slightly longer than broad respectively, Fu_5 and Fu_6 quadrate; each funicular segment with one row of longitudinal sensilla; clava slightly clavate, $2.43\times$ as long as broad, micropilosity only limited to apex of third clava segment. Head in dorsal view $1.89\times$ as wide as long; vertex convex, occipital carina strong; eye length $2\times$ temple length; POL $1.64\times$ OOL.

Head 1.31× as broad as thorax. Mesosoma 1.38 × as long as broad. Pronotum with raised reticulation, pronotal collar slightly narrower than mesoscutum (0.86×); middle length of pronotum almost 1/9 as long as length of mesoscutum; collar not margined anteriorly, posterior margin of collar with a smooth band. Mesoscutum 1.86× as broad as long, with regular and dense reticulation; notauli incomplete and unobvious. Scutellum convex, 1.09× as broad as long, frenal line absent; reticulation same as on mesoscutum but slightly large on posterior part of scutellum. Propodeum (Fig. 32) medially 1/2 as long as scutellum; plica complete; median carina incomplete, occasionally with one or two short longitudinal ridge which interrupted in the middle; nucha short and smooth, separated with middle part of propodeum by a transverse shallow depression; spiracles elongate, 2× as long as broad, separated by the width of spiracles from hind margin of metanotum; area below spiracles with finely reticulation. Fore wing 2.25× as long as broad, without marginal fringe; basal vein with sparse setae, basal cell bare; speculum only stretched to 1/3 base of marginal vein; upper surface of costal cell bare, lower surface with a one complete row of setae and distal 1/3 with some scattered setae; submarginal vein 2.33× as long as marginal vein; marginal vein 1.67× as long as postmarginal vein; stigmal vein as long as postmarginal vein, slightly curved.

Petiole invisible dorsally. Gaster (Fig. 29) long ovate, 1.8× as long as broad, 1.3× as broad as thorax width; surface of each tergite coriaceous; Gt_1 covering 1/3 length of gaster, posterior margin of Gt_1 cambered, median with an obvious hollow; following tergites with posterior margin straight; gaster terminal acute.

Male. Body length 2.1 mm; head and thorax blue-green; antenna light brownish except clava slightly dark, other segments yellow; legs yellow to yellowish brown except coxae concolorous with body; fore wing yellowish brown; gaster brown, with a yellow transverse bright ribbon at 1/3 base of gaster.

Material examined. Holotype: China: ♀, Beijing: Miyun Reservoir, 40.29°N, 116.50°E, 18.VII.1983, ex. pupae of *Dendrolimus tabulaeformis*, leg. Ju-Wen Wu. Paratypes: China: 1♂, 2♀, same data as holotype; 2♀, Beijing: Miyun Reservoir, 20.VII.1983, ex. pupae of *Dendrolimus tabulaeformis*, leg. Ju-Wen Wu; 5♀, Beijing: Huairou, 15.VI.1982, ex. *Illiberis pruni*, leg. Mr. Jin; 1♀, Beijing: Songshan, 26.VIII.1984, ex. *Illiberis pruni*; 4♀, Beijing: Changping, 15.VI.1981, ex. *Locastra muscosalis*, leg. Zhen-Hua Liu; 1♀, Beijing: Yuanmingyuan Imperial Garden, 18.VII.1984, ex. larvae of *Lymantria dispar*, leg. Mu-Zong Cheng; 6♀, Beijing: Yuanmingyuan Imperial Garden, 2.VI.1984, ex. larvae of *Lymantria dispar*, leg. Ding-Xi Liao; 4♂, 5♀, Beijing: Mentougou, late July of 1983, ex. *Prothesia similis xanthocampa*, leg. Sui-Hua Zhao; 1♀, Beijing: Qingbaichang, leg. Ding-Xi Liao.

Etymology. In memory of professor Ding-xi Liao in China.

Hosts. Larvae of *Illiberis pruni* Dyar, *Illiberis nigra* Leech (Lepidoptera: Zygaenidae), *Lymantria dispar* (L.) (Lepidoptera: Noctuoidea) and *Porthesia similis* (Fueszly) (Lepidoptera: Lymantridae), pupae of *Dendrolimus tabulaeformis* Tsai et Liu, *Dendrolimus superans* (Butler) (Lepidoptera: Lasiocampidae), *Illiberis ulmivora* Graeser, *Pseudopanolis flavimacula* Inaba (Lepidoptera: Noctuidae), *Rogas dendrolimi* (Matsumura) (Hymenoptera: Braconidae) and Tenthredinidae sp..

Distribution. China (Liaoning, Beijing, Hebei, Shanxi, Shandong, Gansu, Qinghai).

***Dibrachys maculipennis* Szelényi, 1957, new record to China**

Figs 33–37

Dibrachys maculipennis Szelényi, 1957: 301, 307.

Diagnosis. Body slender (Figs 33, 34), length 2.2–2.3 mm; gaster spindle. Head in frontal view (Fig. 35), 1.13× as wide as high; antennal scrobe very shallow, extending upwards but not reaching anterior ocellus; antennal insertion slightly above lower ocular line; lower face flat; clypeus with transverse striation and lower margin slightly protruding with two blunt teeth. Antennal scape as long as eye height, reaching lower margin of anterior ocellus; length of pedicel and flagellum combined slightly shorter than head width (0.95×); anelli transverse; Fu_1 to Fu_3 slightly long than broad respectively, Fu_4 and Fu_5 quadrate, Fu_6 slightly transverse; clava slightly clavate, 2× as long as broad. Head in dorsal view, 2× as wide as long; occipital carina strong; POL 1.5× OOL. Mesosoma 1.6× as long as broad, with regular reticulation. Propodeum (Fig. 36) with median carina incomplete; plicae distinct anteriorly. Fore wing (Fig. 37) with a yellowish-brown infumation behind marginal vein; submarginal vein 2× as long as marginal vein, marginal vein 1.94× as long as postmarginal vein, 1.5× as long as stigmal vein; stigma vein slightly longer than postmarginal vein (1.1×). Gaster 2× as long as broad, 1.14× as broad as thorax width.

Material examined. China: 2♀, Beijing, V1967; 1♀, Shaanxi: Hangzhou, X.1959, ex. *Smerinthus planus* Walker, leg. Zhe-Min Zheng; 1♀, Zhejiang: Hangzhou, 7.VI.1972, ex. *Apanteles baoris* Wilkinson, leg. Ding-Xi Liao.

Hosts. Recorded hosts of the species were *Megachile rotundata* (Fabricius) (Hymenoptera: Megachilidae) (Peck 1969) and *Hyphantria cunea* (Drury) (Lepidoptera: Erebidae) (Dzhanokmen 1978). Here we newly report *Smerinthus planus* Walker (Lepidoptera: Sphingidae) and *Apanteles baoris* Wilkinson (Hymenopter: Braconidae).

Distribution. China (Beijing, Shaanxi, Zhejiang); Palearctic, Nearctic and Neotropical regions.

***Dibrachys microgastri* (Bouché, 1834)**

Figs 38–41

Diplolepis microgastri Bouché, 1834: 168; neotype female in ZMH by Peters and Baur 2011: 12. Syntypes presumed lost (Graham 1969: 811). Möller 1886: 83; Vidal 2001, 7: 62.

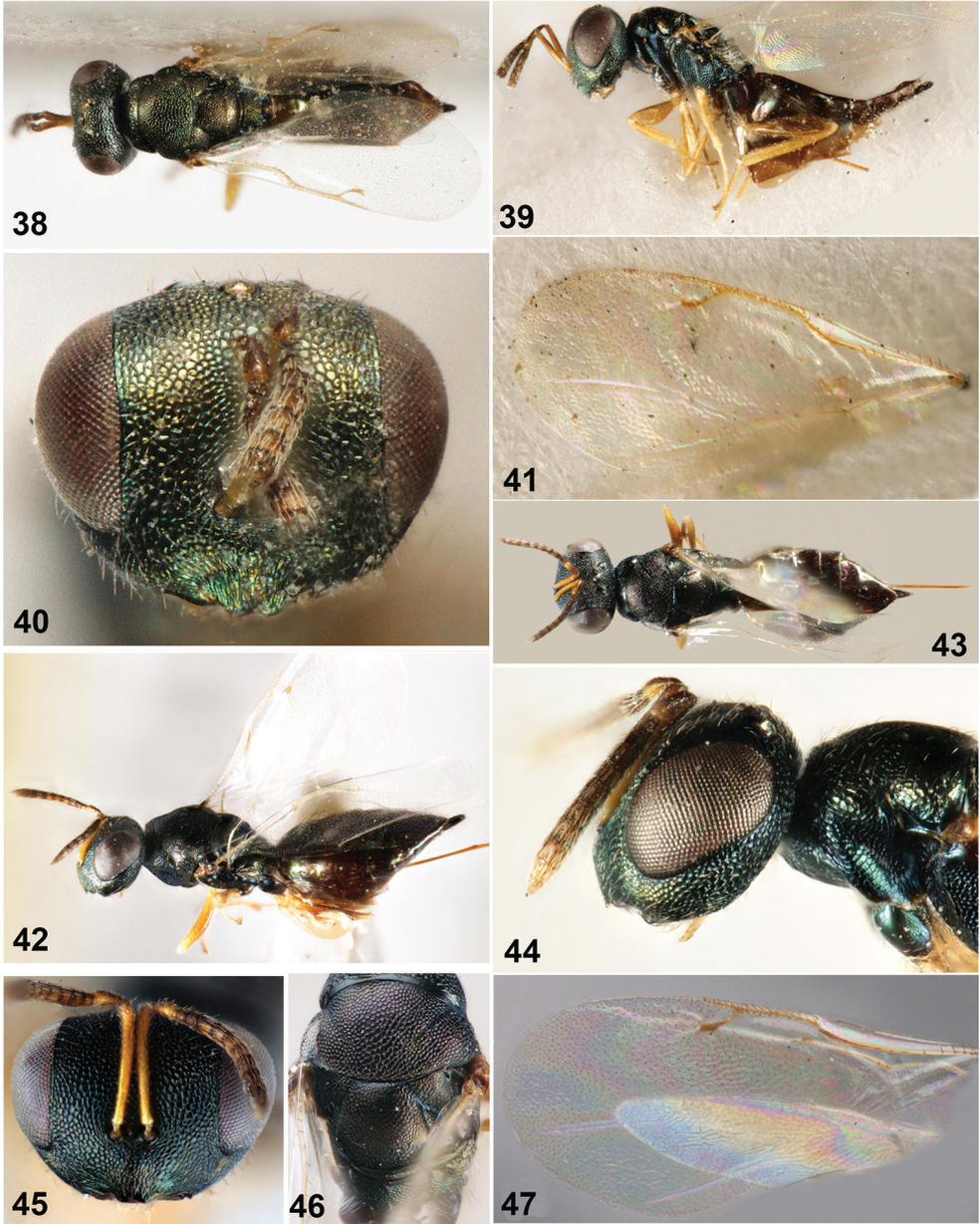
Dibrachys microgastri (Bouché) Peters & Baur, 2011: 18; Vidal 2001: 62.

Synonymy: *Pteromalus cavus* Walker, 1835: 477–478; *Pteromalus decedens* Walker, 1835: 478; *Pteromalus albinervis* Ratzeburg, 1844: 199; *Pteromalus boucheanus* Ratzeburg, 1844: 196; *Pteromalus tenuis* Ratzeburg, 1844: 195; *Pteromalus zelleri* Ratzeburg, 1848: 190; *Pteromalus vesparum* Ratzeburg, 1852: 233; *Cleonymus clisiocampae* Fitch, 1856: 431–432; *Pteromalus boarmiae* Walker in Newman,

1863: 8609, 8610; *Cheiopachus nigro-cyaneus* Norton, 1869: 327; *Eupelmus cereanus* Rondani, 1876: 38, 40; *Pteromalus gelechia* Webster, 1883: 151; *Pteromalus chionobae* Howard, 1889: 1872, 1889; *Arthrolytus apatelae* Ashmead, 1893: 162; *Arthrolytus pimplae* Ashmead, 1894: 339; *Trichomalus truyilloi* Blanchard, 1938: 178; *Tritneptis elegans* Szelenyi, 1981: 400.

Diagnosis. Body slender (Figs 38, 39), length 1.8–2.5 mm; gaster long ovate, spindle-shaped. Head in frontal view (Fig. 40), width $1.21 \times$ height; antennal scrobe extending upwards but not reaching anterior ocellus; antennal insertion placed on lower ocular line; lower face at least slightly convex; sculpture on face larger than on vertex; clypeus with longitudinal striation; lower margin of clypeus slightly protruded, emarginated and with two blunt teeth. Antennal scape slightly shorter than eye height; not reaching lower margin of anterior ocellus; length of pedicel and flagellum shorter than head width; anelli transverse; each funicular segment quadrate except Fu_6 transverse; setae on antenna become an angle with antennal surface. Mesosoma $1.43 \times$ as long as broad; mid lobe of mesoscutum with coarse sculpture. Propodeum with complete plicae and weak median carina. Fore wing (Fig. 41) $1.88 \times$ as long as broad; submarginal vein $2 \times$ as long as marginal vein, marginal vein $1.9\text{--}2.5 \times$ postmarginal vein, postmarginal vein shorter or as most as long as stigmal vein; stigmal vein straight. Gaster $2 \times$ as long as broad, slightly broader than thorax width.

Material examined. China: 3♂, 7♀, Heilongjiang: Hailin, VI.1975, ex. pupae of *Atractodes* sp., leg. Gui-You Zhang; 2♂, 1♀, Heilongjiang: Yichun, 3.VII.1972, ex. Tachinidae sp. on Tortricidae sp., leg. Ding-Xi Liao; 1♂, 8♀ Heilongjiang: Yichun, 13.VII.1962, ex. Tachinidae sp. on *Ptycholomoides aeriferanus* Herrich-Schaffer, leg. Ding-Xi Liao; 2♂, 6♀, Heilongjiang: Yichun, 18.IX.1975, ex. *Ptycholomoides aeriferanus* Herrich-Schaffer, leg. You-Qiao Liu; 4♀, Heilongjiang: Harbin, 3.VII.1962, ex. pupae of *Yponomeuta padella* Linnaeus, leg. Tai-Lu Chen; 5♀, Heilongjiang: Harbin, 4.VII.1962, ex. pupae of flies, leg. Tai-Lu Chen; 1♂, 2♀, Heilongjiang: Harbin, 11.V.1978, ex. pupae of *Anacamptis populella* Clerck, leg. Wen-Min Chen; 2♂, 7♀, Heilongjiang: Harbin, 11.IV.1978, ex. larvae of Aphididae sp., leg. Wen-Min Chen; 11♂, Heilongjiang: Jiamusi, 21.IX.1979, ex. aphids on cabbagecabbage aphid, leg. Ding-Xi Liao; 1♂, 8♀, Heilongjiang: Mishan, VI.1963, ex. *Pyrausta nubilalis* (Hübner), leg. De-Yun Deng; 1♂, 1♀, Heilongjiang: Dailing, 18.IX.1975, ex. *Rhyacionia buoliana*, leg. Ding-Xi Liao; 2♀, Jilin: Siping, VII.1980, ex. *Coleophoridae* sp., leg. Yu-Ying Qiu; 5♀, Liaoning: Liaoyang, 19.VI.1980, ex. larvae of *Lymantria dispar* L., leg. Yu-Bao Zhang and Gui-Zhi Zhang; 3♂, 7♀, Liaoning: Liaoyang, 17.VII.1979, ex. *Musca domestica* (Linnaeus, 1758), leg. Ding-Xi Liao; 1♀, Liaoning: Liaoyang, 10.VI.1979, ex. pupae of *Cnidocampa flavescens* (Walker), leg. Yu-Bao Zhang; 5♀, Liaoning: Jinzhou, vi.1970, ex. *Nephoteryx pivivorella* Matsumura, leg. Bin Liu; 1♂, 1♀, Liaoning: Shenyang, 11.VII.1978, ex. sawfly, leg. Gong-Tian Xu; 1♂, 1♀, Liaoning: Xingcheng, 25.V.1981, leg. Yan-Li Zhao; 4♀, Liaoning: Suizhong, 13.IX.1973, ex. pupae of Tachinidae sp., leg. Shu-Hai Wang; 5♀, Liaoning: Liaoyang, Yuejiadadui, 1979, ex. pupae of *Lymantria*



Figures 38–47. 38–41 *Dibrachys microgastris* (Bouché) 38 Body in dorsal view 39 Body in lateral view 40 Head in frontal view 41 Fore wing 42–47 *Dibrachys qinghaiensis* sp. n., female holotype 42 Body in lateral view 43 Body in dorsal view 44 Head in lateral view 45 Head in frontal view 46 Mesoscutum 47 Fore wing.

dispar (L.) from *Populus* sp., leg. Yu-bao Zhang & Gui-Zhi Zhang; 1♀, Liaoning: Liaoyang, Beiling, 5.IX.1978, ex. *Lymantria dispar* (L.), leg. Gong-Tian Xu; 1♀, Liaoning: Fuxian, 25.VI.1976, ex. *Tortricidae* sp., leg. Ding-Xi Liao; 3♀, Liaon-

ing: Siping, VI.1989, ex. Yponomeutidae sp., leg. Gui-You Zhang; 8♀, Liaoning: Fusong, IX.1953, ex. eggs of *Dendrolimus* sp.; 2♀, Inner Mongolia: Urad Middle Banner, 17.VII.1980, ex. *Anacamptis Populella* Clerck, leg. Xu-Chang Huang; 3♂, Inner Mongolia: Horing, 4.VIII.1981, ex. *Malacosoma* sp., leg. Qiang-Hua Shao; 1♂, 2♀, Inner Mongolia: Baotou, 19.IX.1989, ex. pupae of gelechiid moth, leg. Zhong-Ren Liu; 2♂, 3♀, Inner Mongolia: Baotou, 24.VI.1989, ex. pupae of ichneumon, leg. Zhong-Ren Liu; 1♀, Inner Mongolia: Baotou, 28.vii.1985, ex. Scolytidae sp., Zhong-Ren Liu; 3♂, 7♀, Beijing, vii.1986, ex. pupae of Tachinidae sp., leg. Da-Wei Huang; 1♂, 9♀, Beijing, 21.VIII.1962, ex. Tortricid, leg. Ding-Xi Liao; 5♀, Beijing: Xijiao, 20.IX.1957, ex. pupae of *Stilprotia salicis*, leg. Tai-Lu Chen; 1♀, Beijing: Miyun, 17.VI.1984, ex. *Dendrolimus tabulaeformis*, leg. Da-Wei Huang; 2♂, 6♀, Beijing: Daxing, 1963, ex. obsolete honeycomb, leg. Zong-You Xu; 2♀, Beijing: Yuanmingyuan Imperial Garden, 25.vii.1984, ex. larvae of *Lymantria dispar*, leg. Mu-Zong Cheng; 4♀, Hebei: Zhangbei, VII-XII.1983, ex. *Stilpnotia candida*, leg. Ding-Xi Liao; 8♂, 24♀, Hebei: Zhangbei, 21.VII.1983, larvae of *Stilpnotia candida*, leg. Xing-Jun Li; 9♀, Hebei: Zhangbei, vii.1983, ex. Lymantridae sp., leg. Xing-Jun Li; 1♂, 5♀, Hebei: Zhangbei, ex. pupae of *Gypsonoma minutara*, leg. Jun-Rong Dai; 1♀, Hebei: Fengning, 20.v.1992, ex. *Rogas dendrolimi* (Matsumura), leg. Da-Zhou Wang; 2♂, 3♀, Shanxi: Taiyuan, 6.V.1991, ex. *Eulecanium gigantean*, leg. Hui-Di Zhang; 2♀, Shanxi: Taigu, 8.VII.1979, ex. pupae of Yponomeutidae sp., leg. Zhan-Gui Li; 1♂, 2♀, Shanxi: Taiyuan, 19.vi.1990, ex. pupae of *Yponomeuta polystinellus* Felder, leg. Da-Wei Huang; 1♂, 1♀, Shanxi: Taiyuan, iv.1980, ex. pupae of *Ancylis sativa* Liu, leg. Ci Yu; 3♀, Shanxi: Pingshun, 10.VIII.1978, ex. *Pinus tabuliformis* Carrière; 1♂, 1♀, Shanxi: Taigu, 8.VII.1979, ex. pupae of *Galleria mellonella*, leg. Zhan-Gui Li; 3♀, Shanxi: Taigu, 15.VI.1979, ex. larvae of *Illiberis nigra* Leech, leg. Zhan-Gui Li; 4♀, Shanxi: Taigu, VII.1979, ex. *Illiberis nigra* Leech, leg. Zhan-Gui Li; 1♀, Shanxi: Taigu, VII.1979, ex. *Lithocolletis ringoniella* Mats., leg. Zhan-Gui Li; 2♀, Shanxi: Shuoxian, 24.V.1984, ex. Braconidae on *Coccinella septempunctata*, leg. Yu-Zhi Niu; 3♂, 6♀, Shanxi: Shuoxian, 3-16.VI.1984, ex. Braconidae sp. on *Anacamptis Populella* Clerck, leg. Yu-Zhi Niu; 3♂, 6♀, Shanxi: Shuoxian, 14-18.VI.1984, ex. *Anacamptis Populella* Clerck, leg. Yu-Zhi Niu; 1♂, 2♀, Shanxi: Shuoxian, 20.VI.1983, ex. *Anacamptis Populella* Clerck, leg. Yu-Zhi Niu; 1♂, 7♀, Shanxi: Shuoxian, V-VII.1984, ex. Tachinidae sp. on *Anacamptis Populella* Clerck, leg. Yu-Zhi Niu; 1♀, Shangdong: Weihai, 3.VI.1958, leg. Jin-Long Mao; 1♀, Shangdong: Fushan, 26.X.1958, leg. Jin-Long Mao; 1♂, 4♀, Henan: Anyang, 20.V.1956, ex. *Pectinophora gossypiella* (Saunders), leg. Ding-Xi Liao; 3♀, Henan: Zhengzhou, 10.IX.1972, ex. larvae of Olethreutidae sp., leg. Ding-Xi Liao; 1♀, Shaanxi: Xianyang, 6.III.1975, ex. pupae of *Earias cupreoviridis* Walker, leg. Ding-Xi Liao; 2♂, 6♀, Ningxia: Luhuatou, 29.V.1982, ex. *Ypsolopha vittellus* (Linnaeus), leg. Ding-Xi Liao; 3♀, Ningxia: Zhongweis-Shapotou, 15.VI.1981, leg. Ding-Xi Liao; 3♀, Ningxia: Yinchuang, 12.VI.1974, ex. pupae of Syrphidae sp. on *Ulmus pumila* L.; 1♂, 9♀, Gansu: Pingliang, VII.1966, ex. Tachinidae sp. & *Illiberis pruni*, leg. Shou-Min Liu; 3♀12♂, Gansu: Pingli-

ang, VII. 1966, ex. *Illiberis pruni*, leg. Shou-Min Liu; 4♂, 22♀, Xinjiang: Altay, VI.1979, ex. larvae of *Gelechia pinguinella* Trietschke, leg. Jun-Wen Xia; 5♀, Xinjiang: Korla, collecting time unknown, ex. eggs of *Macroglossum corythus luteata* (Butler), leg. Tai-Lu Chen; 3♀, Xinjiang: Urumchi, 12.VII.1980, ex. pest on *Salix* sp., leg. Jiu-Xiong Bai; 1♂, 5♀, Jiangsu: Nanjing, 4.VII.1963, leg. Ding-Xi Liao; 2♂, 5♀, Shanghai: Pudong New District, III.1972, ex. cottonseed; 4♀, Shanghai: Minhang, XII.1979, ex. *Apanteles glomeratus* (L.), leg. Ji-Long He; 6♀, Anhui: Dangshan, 7.XI.1975, ex. *Pyrausta nubilalis* (Hubern) & *Chilo infuscatellus* (Snellen), leg. Ding-Xi Liao; 12♀, Anhui: Huangfu Mountain, 19.VI.1965, ex. larvae of Lepidoptera, leg. Ding-Xi Liao; 1♂, 4♀, Zhejiang: Hangzhou, X.1954, ex. *Pectinophora gossypiella* (Saunders); 2♂, 6♀, Zhejiang: Hangzhou, i.1963, leg. Ding-Xi Liao; 5♂, 2♀, Zhejiang: Hangzhou, I.1963, ex. *Pectinophora gossypiella* (Saunders), leg. Cui Hu; 1♂, 2♀, Hubei: Hong'an, V.1978, ex. pupae of *Dendrolimus punctatus* Walker, leg. Ding-Xi Liao; 6♀, Hunan: Changsha, 20.III.1984, ex. *Chilo suppressalis* (Walker), leg. Ding-Xi Liao; 1♂, 9♀, Hunan: Liuyang, 24.IX.1979, ex. eggs of *Dendrolimus* sp., leg. Ding-Xi Liao; 1♀, Hunan, collecting time unknown, leg. Xin-Wang Tong; 1♀, Yunnan: Kunming, 17.V.1967, ex. larvae of *Macrocentrus* sp. leg. Jing-Liang Qi; 1♀, Yunnan: Zhaotong, 5.IV.1973, ex. *Chilo suppressalis* (Walker), leg. Ding-Xi Liao; 1♂, 1♀, Tibet: Lhasa, 3650m, 27.VIII.2001, leg. Chao-Dong Zhu. 2♀, Sk. Ahus, 15.IV.1979. leg. K. J. Hedqvist, *Dibrachy cavus* (Walker), det. K. J. Hedqvist; N. Zealand: 1♀, Lincoln, nr Christchurch RT., 1988, B.J. Donovan. Cult. *Vespula germanica*, *Dibrachys boarmiae* (Walker), det. Z. Bouček, 1988.

Hosts. Hosts of *D. microgastri* has been widely recorded, and the primary parasite has been recorded more than 240 species from 45 families of seven insects orders and also recorded two species of Arachnida (Noyes 2002). In our study, the species was parasitic on Coleoptera (Scolytidae sp.), Diptera (Tachinidae sp. on Tortricidae sp., Tachinidae sp. on *Anacamptis Populella* Clerck, pupae of Tachinidae sp. and Syrphidae sp. on *Ulmus pumila* L.), Hemiptera (aphids on cabbage), Hymenoptera (*Apanteles glomeratus* (L.), Braconidae sp. on *Coccinella septempunctata*, larvae of *Macrocentrus* sp., pupae of *Gelis* sp. and Lepidoptera (*Chilo infuscatellus* (Snellen), *Chilo suppressalis* (Walker), *Lymantria dispar* (L.), *Malacosoma* sp., *Nephopteryx pivorella* (Matsumura), *Pectinophora gossypiella* (Saunders), *Ptycholomoides aeriferanus* (Herrich-Schäffer), *Pyrausta nubilalis* (Hübner), *Ypsolopha vittellus* (Linnaeus), eggs of *Dendrolimus* sp., larvae of *Gelechia pinguinella* Trietschke and Olethreutidae sp., pupae of *Anacamptis populella* Clerck, pupae of *Cnidocampa flavescens* (Walker), pupae of *Dendrolimus punctatus* (Walker), pupae of *Earias cupreoviridis* Walker, pupae of *Stilprotia salicis* (L.), pupae of *Yponomeuta padella* Linnaeus and so on).

Distribution. China (Heilongjiang, Jilin, Liaoning, Inner Mongolia, Beijing, Hebei, Shanxi, Shangdong, Henan, Shaanxi, Ningxia, Gansu, Xinjiang, Jiangsu, Shanghai, Anhui, Zhejiang, Hubei, Hunan, Yunnan, Tibet); widespread world-wide distribution (Noyes, 2016).

***Dibrachys qinghaiensis* Jiao & Xiao, sp. n.**

<http://zoobank.org/2C61C736-41D3-4EB0-8C0B-F3218A96B622>

Figs 42–47

Diagnosis. The new species belongs to *Dibrachys* s. str., and the mainly differences with *Dibrachys microgastri* (Bouché) are as follows: antenna of *D. qinghaiensis* sp. n. slender, each funicular segment at least slightly longer than its broad; antennal scape as long as eye height, and nearly reaching lower margin of anterior ocellus; but *D. microgastri* (Bouché) at least with several transverse funicular segment in distal of antenna, antennal scape distinctly shorter than eye height, and not reaching lower margin of anterior ocellus.

Description. Holotype. *Female*. Body (Figs 42, 43) length 2.2 mm. Head and mesosoma dark green, with brown gloss and metallic reflection; gaster dark brown. Antenna with scape and pedicel yellowish brown, but brownish in dorsum, other segments of antenna dark brown; mandible yellowish brown and margin of teeth brownish; legs yellowish brown except coxae concolorous with body; fore wing hyaline, wing venation light yellow.

Head in frontal view, 1.27× as wide as high (Fig. 45); frons with very dense reticulation; antennal scrobe with rather large reticulation; lower face flat, with densely transverse striation except lower edge of clypeus smooth; eye height 0.64× head height, eyes separated by 1.23× eye height; scrobe shallow, extending upwards but not reaching anterior ocellus. Antennal insertion slightly above lower ocular line, distance from upper margin of torulus to anterior ocellus 1.58× distance from lower margin of torulus to lower margin of clypeus; clypeal margin protruded, emarginate in middle with two small blunt teeth; oral fossa width 0.59× head width. Head in lateral view (Fig. 44) with malar sulcus inconspicuous, eye height 2.2× malar space. Antennal scape as long as eye height, nearly reaching lower margin of anterior ocellus; length of pedicel and flagellum combined shorter than head width (0.85×); pedicel in lateral view 3× as long as broad; anelli transverse; each funicular segment slightly longer than broad; each funicular segment with one row of longitudinal sensilla; clava slightly clavate, 2.57× as long as broad, micropilosity only limited to apex of third clava segment. Head in dorsal view 2× as wide as long, vertex convex, occipital carina strong; eye length 1.87× temple; POL 1.49× OOL.

Head width 1.31× as broad as thorax. Mesosoma 1.43× as long as broad. Pronotum with coarse reticulation, 0.87× as broad as thorax; pronotum with middle length 0.23× as long as mesoscutum, collar subhorizontal and not margined, posterior margin smooth. Mesoscutum 2× as broad as long, with finely dense reticulation (Fig. 46), posterior reticulation larger than anterior reticulation; notauli incomplete but conspicuous anteriorly. Scutellum flat, as long as broad, frenal line absent; finely reticulate. Propodeum medially 0.43× as long as scutellum, central area flat and with regular reticulation; plicae complete and parallel anteriorly, separated by 1.82× medial length of propodeum; median carina complete; propodeum with short, convex nucha; spiracles elongate, 2× as long as broad, separated by the width of spiracles from hind margin of metanotum; area below spiracles with conspicuous and deep reticulation. Fore wing (Fig. 47) 2.38× as long as broad, without marginal fringe; upper surface densely pubes-

cent; basal vein with sparse setae, basal cell bare, speculum only stretched to the base of marginal vein; upper surface of costal cell bare, lower surface with one complete row of setae and distal 1/3 with two rows of short setae; submarginal vein 2.37× as long as marginal vein; marginal vein 2.64× as long as stigmal vein; postmarginal vein as long as marginal vein; stigmal vein slightly curved.

Petiole invisible dorsally. Gaster long ovate, 2× as long as broad; 0.89× as broad as thorax width, 1.33× as long as length of mesosoma; surface of each tergite coriaceous; Gt_1 covering 1/3 of gaster, posterior margin straight and with small hollow in middle; posterior margin of other tergites straight; terminal acute.

Male. Body length 1.3–1.9 mm, head and mesosoma black, with yellow-green shine; antennae yellow; legs yellow except coxae concolorous with body. Antennae with long hair, each funicular segment longer than its broad, with long hair on antenna. Gaster ovate, with an oval pale spot between Gt_1 and Gt_2 .

Material examined. Holotype. China: ♀, Qinghai: Golmud, Guolemude, 2880m, 36.26°N, 94.53°E, 14.IX.2001, leg. Chao-Dong Zhu. Paratype. China: 5♂, 4♀, same data as holotype; 2♀, Qinghai: Delhi, Baingoin, 2900m, 16.IX.2001, leg. Chao-Dong Zhu; 1♀, Qinghai: Qilian, 2790m, 19.IX.2001, leg. Chao-Dong Zhu; 2♀, Qinghai: Dulan, Xiangride, 10.VI.1997, leg. Chao-Dong Zhu; 4♀, Qinghai: Xining, 3–4.VI.1997, leg. Chao-Dong Zhu; 2♀, Qinghai: Tongren, Mailin, 14.VI.1997, leg. Chao-Dong Zhu.

Etymology. The specific name is consist of the spelling of the type locality “qinghai” and the suffix “-ensis” represent source.

Hosts. Unknown.

Distribution. China (Qinghai, Yunnan).

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