A new species of *Metallus* Forbes (Hymenoptera: Tenthredinidae) from China with a key to world species

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Abstract: A new leafmining sawfly species is described from China: *Metallus nanlingicus* Wei **sp. nov.** The mitochondrial genome of the new species is also sequenced. Based on the known *COI* data, the phylogenetic relationship of some species of *Metallus* is reconstructed. The preliminary result shows that *M. nanlingicus* Wei **sp. nov.** is close to *M. mai* Wei, 1994 from China and the lineage of *M. nanlingicus* + *M. mai* is the sister group of *M. albipes* (Cameron, 1875) from Europe. Based on this phylogenetic analysis some misidentified samples in NCBI were revealed. A key to known species of *Metallus* is also provided.

Key words: Tenthredinomorpha; Fenusinae; leafmining sawfly; taxonomy; mitochondrial genome

中国昧潜叶蜂属一新种暨世界种类检索表（膜翅目：叶蜂科）

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摘要：记述中国潜叶蜂 1 新种：南岭昧潜叶蜂 *Metallus nanlingicus* Wei **sp. nov.**，测序分析了该种的线粒体基因组，并基于已知昧潜叶蜂属 *COI* 数据构建了系统发育关系。初步分析结果表明，南岭昧潜叶蜂与马氏昧潜叶蜂关系较近，*M. nanlingicus* + *M. mai* 支系是 *M. albipes* 的姊妹群。基于系统发育关系分析，指出 NCBI 系统中部分昧潜叶蜂属的 *COI* 数据存在样本鉴定错误。文中还编制了昧潜叶蜂属世界已知种类检索表。

关键词：叶蜂亚目；潜叶蜂亚科；潜叶蜂；分类；线粒体基因组

Introduction

Metallus Forbes, 1885 is the largest genus in the subfamily Fenusinae of Tenthredinidae with 19 known species and 1 subspecies (Malaise 1964; Smith 1981, 1988; Lee & Ryu 1996; Nie & Wei 1998; Wei & Xiao 2005; Hara & Ibuki 2023). North America and Europe are both home to four species, one of which is common to both regions (Eiseman & Smith 2017). The remaining 13 species and 1 subspecies of this genus are endemic to East Asia, and one European species has also been reported in Japan.

The southeastern region of China has the largest specific diversity of *Metallus*. In total, 10 species have been reported from this region (Wei 1994; Xiao *et al.* 1997; Nie & Wei 1998; Wei *et al.* 2003; Wei & Xiao 2005) and there are at least 5 additional undescribed species in the collection of the Asian Sawfly Museum, Nanchang, China. Here we describe one new

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species, and the mitochondrial genome of the species is also briefly reported.

Material and methods

Specimens were examined with a Motic-SMZ-171 stereomicroscope. Images of adults were taken with a Nikon D700 digital camera and a Leica Z16APO microscope. Images were focus-stacked using Helicon Focus (HeliconSoft, Kharkiv, Ukraine) and further processed with Adobe Photoshop CS 23. The terminology of genitalia follows Ross (1945) and terminology of wing venation follows Niu and Wei (2010) except for m-cu being replaced by 1m-cu, and stigma by pterostigma.

For the methods and procedure of the mitogenome sequencing, annotation and analyses, and the phylogenetic analyses in detail, see Niu *et al*. (2021).

The types of these new species are deposited in the Asian Sawfly Museum, Nanchang, China (ASMN).

Taxonomy

***Metallus nanlingicus* Wei sp. nov.** (Fig. 1)

Female length 4–5 mm. Body black, palpi largely white; legs yellowish white, fore coxa and trochanter entirely, basal 2/3 of middle and hind coxae, four and middle femora largely blackish brown; fore wing evenly and strongly infuscate, veins and stigma blackish brown, hind wing subhyaline; body hairs pale brown, hairs on dorsum of head and of thorax dark brown.

Body strongly smooth; frons with minute and very sparse punctures, punctures on mesonotum small and sparse but distinct. Hairs on dorsum of head and mesonotum not longer than diameter of ocellus. Anterior margin of clypeus faintly and roundly incised, almost truncate; malar space distinct but linear; middle fovea large and shallow, lateral fovea quite small, deep and round; inner margins of eyes weakly bent, distinctly convergent downwards, distance between lower corner of eyes slightly longer than longest axis of eye (Fig. 1B); interocellar furrow punctiform, postocellar area feebly elevated, 2 times broader than long anteriorly and 2.7 times broader than long posteriorly, without middle furrow; lateral furrows deep and strongly divergent backwards, longer than diameter of lateral ocellus; head behind eyes strongly narrowing (Fig. 1A); antenna about 1.7 times head breadth, 3rd antennomere 1.2 times as long as 4th one, fifth antennomere about 2.8 times as long as broad, last antennomere 1.3 times as long as penult antennomere, about 3 times as long as broad, distinctly tapering toward apex (Fig. 1F). Hind basitarsus distinctly longer than 3 following tarsomeres together. Fore wing with first part of vein Rs largely vestigial as a white line, vein 1M strongly but roundly bent at basal 2/5 and oblique to vein M+Cu, cu-a meeting cell 1M slightly beyond middle, 2r beyond middle of cell 2Rs, apical part of vein 2A vestigial; cell R1 in hind wing close, anal petiole 1.1–1.2 times as long as vein cu-a (Fig. 1D). Middle membranous area of first tergum small and triangular; apical sheath narrowed toward apex, dorsal apical corner narrowly roundish (Fig. 1C); lancet with 14 oblique serrulae (Fig. 1G), third serrula as long as convex membranous part between third and fourth serrulae, with subbasal teeth minute and

indistinct (Fig. 1H); apical serrulae not touching each other (Fig. 1I).

Male. Body length 3.8–4.0 mm. Color and structure similar to female except for antennal flagellum brownish and strongly compressed (Fig. 1E), hind tibia stout, metabasitarsus as long as following 3 tarsomeres together, subgenital plate as long as broad with apical margin broadly convex; parapenis higher than broad, harpe longer than broad with long spines, apical margin oblique (Fig. 1K); penis valve as shown in Fig. 1J, valviceps broad, ventral margin broadly truncate.

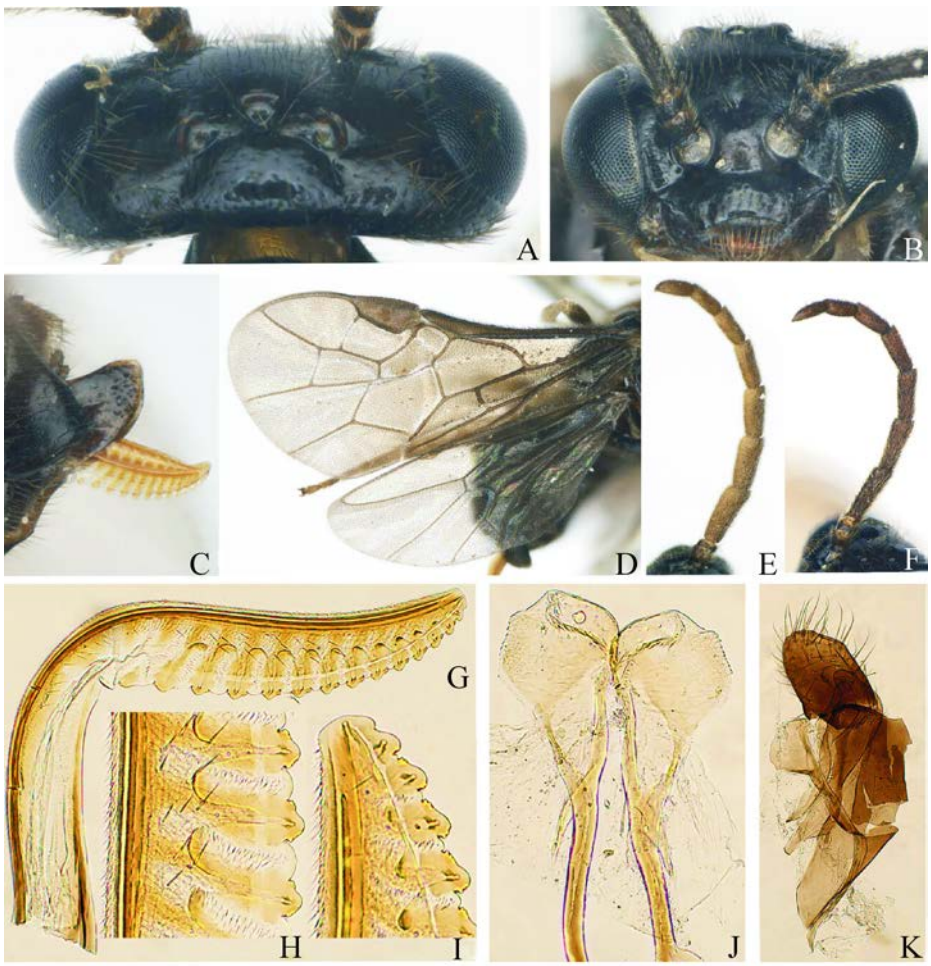

Figure 1. *Metallus nanlingicus* Wei **sp. nov.** paratype. A, B. Female head, dorsal and frontal views; C. Ovipositor sheath, lateral view; D. Female wings; E. Male antenna; F. Female antenna; G. Lancet; H. Third to fifth serrulae; I. Apical serrulae; J. Penis valves; K. Gonoforcep.

Holotype. ♀, **China**, Hunan, Mt. Mangshan, Guizizhai, 112º56.111´E, 24º57.002´N, alt. 1,240 m, 11-IV-2007, Qun YOU leg. **Paratypes**. 2♀, **China**, Hunan, Mt. Mangshan, Datangkeng, 112º48.138´E, 24º57.413´N, alt. 1,090 m, 23-IV-2006, Hu ZHOU leg.; 1♀, **China**, Hunan, Mt. Mangshan, Datangkeng, 112º52.393´E, 24º56.318´N, alt. 1,030 m, 22-IV-2006, Hu ZHOU leg.; 4♀1♂, **China**, Hunan, Daoxian County, Houzi'ao, 111º21.905´E,

25º30.879´N, alt. 806 m, 22-IV-2008, Qun YOU & Fu ZHAO legs; 1♀, **China**, Hunan, Daoxian County, Qingliyuan, Xibian, 111º21.564´E, 25º29.200´N, alt. 500 m, 23-IV-2008, Tianming SU leg.; 1♀, **China**, Hunan, Dao County, Qingliyuan, 111º23.158´E, 25º29.540´N, alt. ??? m, 23-IV-2008, Tianming SU leg.; 1♀, **China**, Guangdong, Shixing County, Mt. Chebaling, 24º40′N, 114º09′E, alt. 400 m, 13-IV-2007, Meicai WEI leg.; 1♀, **China**, Guangdong, Ruyuan County, Mt. Nanling, Qinshuigu, 24º54.990′N, 113º02.093′E, alt. 824 m, 18-VI-2007, Qun YOU leg.; 1♀, **China**, Guangdong, Shixing County, Mt. Chebaling, 24º43.382′N, 114º15.383′E, alt. 400 m, 20-VI-2007, Hanlan FEI legs.; 1♂, **China**, Jiangxi, Pingxiang, Luxi County, 03-IV-2004, Meicai WEI leg.; 1♂, **China**, Guizhou, Chishui, Mt. Jinsha, alt. 500 m, 23-IX-2000, Wei XIAO leg.; 2♀, **China**, Hunan, Yanling County, Mt. Taoyuandong, alt. 900–1,000 m, 23-IV-1999, Meicai WEI leg.; 1♀, **China**, Hunan, Shimen County, Mt. Huping, alt. 500 m, 02-VI-2002, Jigang JIANG leg.

Other specimen examined. 1♀, **China**, Hunan, Mt. Jintong, Yaorenping, 26º15′5″N, 110º30′18″E, alt. 898 m, 20–24-V-2021, Wei XIAO leg.

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

Remarks. This new species is similar to *Metallus nigritarsus* Nie & Wei, 1998 from Zhejiang and Fujian of China, but differs from the latter by the hind femur and hind tarsus yellowish white, the length of cypsella between the third and fourth serrulae as long as the third serrula, the valviceps of the penis valve evenly narrowed toward ergot, ventral margin broadly truncate. In *M. nigritarsus* the hind femur and hind tarsus largely blackish brown, the length of cypsella between the third and fourth serrulae much shorter than the third serrula, the valviceps of the penis valve not evenly narrowed toward ergot with ventral margin subtriangularly convex.

The mitochondrial genome of *Metallus nanlingicus* Wei sp. nov.

Using Getorganelle (Jin *et al*. 2020), a 10 GB raw sequence was assembled, resulting in a complete sequence of 15,555 bp with 37 genes. The accuracy of this sequence was validated against reference sequences *Fenusella taianensis* (OR730573) (unpublished), *Kaliofenusa tangi* (OP429198) (unpublished), and *Kaliofenusa maculoscapa* (OP429196) (unpublished). Ultimately, we obtained a circular sequence of *M. nanlingicus* (OR725470) with a total length of 16,307 bp, comprising 13 protein-coding genes (PCGs), 2 rRNA genes, and 22 tRNA genes. Unlike the putative ancestral insect gene arrangement of *trnI*(+)-*trnQ*(-)-*trnM*(+) (Boore 1999), the sequence has been rearranged to *trnM*(+)-*trnQ*(+)-*trnI*(+), which is consistent with the gene arrangement observed in the only other reported species of the *Metallus* genus, *M. mai* (MW255941) (Niu *et al*. 2021). The overall AT content of the mitochondrial genome is 81.8% (44.0% A, 11.2% C, 7.0% G, 37.8% T), indicating an AT bias. The 13 PCGs feature three types of start codons: ATG (*nad2*, *atp6*, *cox3*, *nad4*, *cob* and *nad1*), ATT (*cox1*, *cox2*, *atp8* and *nad3*), and ATA (*nad5*, *nad4l* and *nad6*). With the exception of *nad4*, which terminates with a T codon, the other 12 PCGs all terminate with TAA.

The phylogeny of *Metallus* based on known *COI* data

Within *Metallus*, no mitogenome data has been reported before this paper. Based on 48 *COI* data of 6 *Metallus* species, a phylogenetic tree was reconstructed (Fig. 2). The new species and 5 known species of *Metallus* form three monophyletic branches. *M. nanlingicus* is

close to *M. mai* from China and these two species form a sister group of *M. albipes*. *M. lanceolatus* is the sister species of *M. pumilus*. *M. rohweri*, a peculiar American species with strange penis valve, is the sister group of other *Metallus* species.

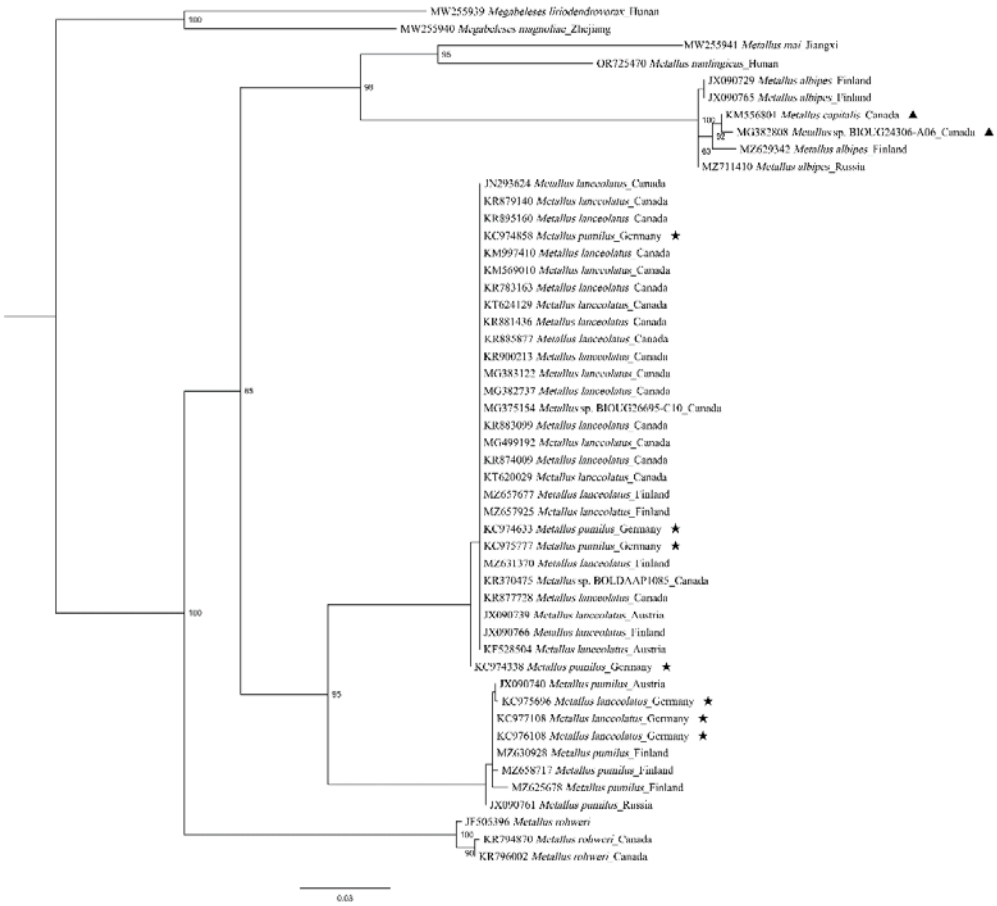

Figure 2. An IQ-tree based on known *COI* sequences of *Metallus* spp. ★, Sample misidentified; ▲, Possible synonym.

Based on the two *COI* data, *M. capitalis* from Canada seems to be an ingroup of *M. albipes*, though more material and mitogenome data are needed to examine the possible synonymy. Except for a few species, most of *Metallus* species are quite similar in body color. The identification of *Metallus* species is a challenging work for common biological researchers. The *COI* data, even the mitogenome data, are important for the correct identification of species. This study on the phylogeny of *Metallus* species shows that the identifications of some *Metallus* species within NCBI are not correct (Fig. 2, samples with black pentagram mark).

Key to known species of *Metallus* worldwide

1. Hind femur and tarsus yellow or white, dorsum of hind femur or hind tarsus sometimes slightly darkened ‥ …… 2

-. Hind femur or hind tarsus black to brownish black…… 17

2. Posterior margin of pronotum and tegula orange to white, mesonotum and metanotum orange, abdomen orange with apical segment lightly infuscate, sheath orange; lancet with 12 serrulae, cypsella acutely incised. North America ····· *M. ochreus* Smith
-. Pronotum, tegulae, abdomen and sheath always black to brownish black ····· 3
3. Lancet with 11 oblique serrulae far apart from each other, membranous part between basal several serrulae flat ····· 4
-. Lancet with 13–16 serrulae, membranous part between basal several serrulae short, usually distinctly convex; penis valve without apical filament ····· 6
4. Body length 2.6–2.8 mm; distance between serrulae 4–5 shorter than serrula 4 with cypsella roundly convex; penis valve with a long and slender filament. Host unknown. China (Zhejiang, Hunan) ····· ····· *M. minutus* Nie & Wei
-. Body length 4 mm; distance between serrulae 4–5 longer than serrula 4 with cypsella flat; valviceps of penis valve broad without a long and slender filament. Host: *Geum.* Holarctic ····· 5
5. Labrum yellow white; base of coxa brown, cercus pale brown. Europe and North America ····· ····· *M. lanceolatus* (Thomson)
-. Labrum dark brown with base blackish; coxa largely black; cerci dark brown. Japan ····· ····· *M. lanceolatus japonicus* Hara & Ibuki
6. North American species; lancet with 13 serrulae, all cypsellae flat ····· *M. capitalis* (Norton)
-. Palaearctic or Oriental species; lancet with 14–16 serrulae, cypsellae convex ····· 7
7. Postocellar area 1.5 times as wide as long, lateral furrows linear, long and sharp; serrulae wider than long and less close to each other. China (Sichuan) ····· *M. wangi* Wei
-. Lateral furrows short and broad, not linear, postocellar area more than 1.5 times as wide as long; serrulae as long as wide or close together ····· 8
8. Middle fovea absent, third antennomere 2.7 times as long as broad; penis valve with a distinct apical horn. Male. China (Hunan, Guizhou) ····· *M. jingfanus* Nie & Wei
-. Middle fovea at least shallowly defined; third antennomere in male less than 2.5 times as long as broad; penis valve without apical horn ····· 9
9. Scape at least largely pale brown ····· 10
-. Scape black; postocellar area 1.8–2 times (♀) or 2.5 (♂) times in male as wide as long ····· 12
10. Postocellar area 2.5 (♀) or 3 (♂) times as broad as long; body length 4–4.5 mm; hind tibia and hind tarsus entirely yellow white; third antennomere clearly longer than fourth one; lancet with 13 serrulae. China (Sichuan, Guizhou) ····· *M. sichuanensis* Wei
-. Postocellar area 2 times as broad as long; body length 2.5–4 mm; apex of hind tibia and hind tarsus darkened or lancet with 16 serrulae ····· 11
11. North Palaearctic species; third antennomere clearly longer than fourth one; apex of hind tibia and hind tarsus darkened; lancet unknown. Europe; Japan ····· *M. albipes* (Cameron) [part]
-. Nepal species; third antennomere about as long as fourth antennomere; hind tibia and hind tarsus entirely pale; lancet with 16 serrulae ····· *M. nepalensis* Smith
12. Fore and middle femora distinctly blackened. China (Jiangxi, Hunan, Guangdong, Guizhou) ····· ····· *M. nanlingicus* Wei **sp. nov.** [part]
-. Middle femur yellowish white ····· 13
13. Apex of sheath round, dorsal corner not acute; lancet with 16 serrulae, subapical serrulae close to each other and strongly convex, hardly oblique, both sides with many fine teeth. China (Hunan, Guizhou) ····· ····· *M. theca* Wei
-. Apex of sheath triangular, dorsal corner acute; lancet with 14–15 serrulae, subapical serrulae strongly

oblique with few fine teeth laterally ····· 14
14. Body length 5–6 mm; fore femur entirely yellowish white; postocellar area 2 times as broad as long ···· 15
-. Body length 3–4 mm; dorsum of fore femur blackish brown; postocellar area 1.8 or 3 times as broad as long ····· 16
15. Vein 1M in fore wing strongly bent at basal 2/5, apical 3/5 close to vein Sc; length of anal petiole of hind wing 1.3 times breadth of anal cell; anterior margin of clypeus truncate; postocellar area without middle furrow, lateral furrows strongly divergent backwards. China (Hunan) ····· *M. melanopterus* Wei
-. Vein 1M in fore wing strongly bent at basal 1/5, apical 4/5 not close to vein Sc; length of anal petiole of hind wing about 2 times breadth of anal cell; anterior margin of clypeus roundly incised; postocellar area with shallow middle furrow, lateral furrows weakly divergent backwards. Europe ····· *M. beckeri* (Konow)
16. Body shorter than 3 mm; middle serrulae as long as wide and far apart from each other, male antenna feebly compressed, third antennomere slightly broader than apical antennomere; female postocellar area 3 times as broad as long. China (Guizhou, Yunnan) ····· *M. bui* Nie & Wei
-. Body longer than 3 mm; middle serrulae close together, and each serrula much broader than long; male antenna strongly compressed, 3rd antennomere much broader than apical antennomere; female postocellar area 1.8 times as broad as long. China (Zhejiang, Fujian, Jiangxi) ····· *M. mai* Wei
17. Middle and hind trochanters, hind femur yellowish white ····· 18
-. All trochanters blackish brown or entirely pale; hind femur largely blackish brown ····· 20
18. Fore trochanter and hind tarsus largely blackish brown; postocellar area 2 times as broad as long; middle serrulae close together and with 4 posterior fine teeth; penis valve ventrally distinctly convex. China (Zhejiang) ····· *M. nigritarsus* Nie & Wei
-. Fore trochanter yellowish white, hind tarsus yellowish white or slightly darkened; postocellar area 2.5–3 times as broad as long ····· 19
19. All femora white; postocellar area 3 times as broad as long. Europe ····· *M. albipes* (Cameron) [part]
-. Dorsum of fore and middle femora blackish brown; postocellar area 2.5 times as broad as long. China (Jiangxi, Hunan, Guangdong, Guizhou) ····· *M. nanlingicus* Wei **sp. nov.** [part]
20. Apex of cell R1 in hind wing open; middle fovea shallow and obscure. East Asian species ····· 21
-. Apex of cell R1 in hind wing close; middle fovea distinct ····· 23
21. Postocellar area 1.7–2.5 times as broad as long; anal petiole of hind wing slightly longer than vein cu-a; metabasitarsus largely pale brown. Korean species ····· 22
-. Postocellar area 3 times as broad as long; metabasitarsus largely blackish brown; anal petiole of hind wing distinctly longer than vein cu-a. China (Guangxi) ····· *M. nigrofemoratus* Wei
22. Body length 3 mm; postocellar area 2.5 times as broad as long; third antennomere 1.2 times as long as antennomere 4; lancet with 12 serrulae. South Korea ····· *M. adamantis* Lee & Ryu
-. Body length 4 mm; postocellar area 1.7 times as broad as long; third antennomere 1.33 times as long as antennomere 4; lancet unknown. South Korea ····· *M. satoi* Smith
23. European or Asian species; thorax always black; lancet narrow with distinct cypsellae; valviceps of penis valve not slender and without long apical process ····· 24
-. North American species; female thorax partly rufous or black; lancet broad and without distinct cypsella; valviceps of penis valve not slender and without apical process ····· *M. rohweri* MacGillivray
24. European species; anal petiole of hind wing much longer than breadth of anal cell; lateral furrows of postocellar area strongly divergent backwards ····· *M. pumilus* (Klug)
-. South Asian species; anal petiole of hind wing slightly longer than breadth of anal cell; lateral furrows of postocellar area weakly divergent backwards. North Myanmar ····· *M. compressicornis* Malaise

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