

DNA barcoding for molecular identification of the genus *Oxyscelio* (Hymenoptera, Scelionidae) from southern China, with descriptions of five new species

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Academic editor: Elijah Talamas | Received 21 July 2021 | Accepted 30 August 2021 | Published 23 December 2021

<http://zoobank.org/CACE04D1-17FF-46DE-84CB-A8302CF3EE8E>

Citation: Mo W-h, Chen H-y, Pang H, Liu J-x (2021) DNA barcoding for molecular identification of the genus *Oxyscelio* (Hymenoptera, Scelionidae) from southern China, with descriptions of five new species. In: Lahey Z, Talamas E (Eds) Advances in the Systematics of Platygastroidea III. Journal of Hymenoptera Research 87: 613–633. <https://doi.org/10.3897/jhr.87.71912>

Abstract

Species of the genus *Oxyscelio* Kieffer are egg parasitoids of Orthoptera. The genus is relatively diverse in China, with 34 described species. Some species of the genus are extremely morphologically similar and difficult to identify, especially in males. In this study, DNA barcoding based on sequences of the *COI* gene was used to discriminate *Oxyscelio* species from southern China. In total, 49 *COI* sequences belonging to 22 morphospecies were obtained. The *COI* sequences worked well for the identification of all the studied species, with intraspecific genetic distances ranging between 0 and 4.3%, while interspecific distances ranged between 7.1% and 19%. Based on both morphological and molecular analyses, five species are described as new: *O. amalocarina* Mo & Chen, **sp. nov.**, *O. apheles* Mo & Chen, **sp. nov.**, *O. latheticus* Mo & Chen, **sp. nov.**, *O. stenos* Mo & Chen, **sp. nov.**, and *O. striae* Mo & Chen, **sp. nov.**

Keywords

Egg parasitoid, integrated taxonomy, molecular species delimitation, Platygastroidea

Introduction

Oxyscelio Kieffer is a large genus of parasitoids attacking the eggs of Orthoptera (Kononova and Fursov 2007), with 186 species having been described from the Indo-Malayan, Palearctic, Australian, Pacific and African regions (Burks et al. 2013a, 2013b, 2016; Mo et al. 2020). Species of the genus are easily recognized by the following combination of characters: submarginal vein of forewing distant from wing margin, marginal vein very short, postmarginal vein absent or very short, vertex rounded posteriorly, metascutellum distinct and usually excavate dorsally, and many species have a pronounced frontal depression on the head which is often rimmed by a carina (Burks et al. 2013a). However, some species within the genus are extremely similar morphologically and difficult to identify. In fact, some similar species were suspected to be the same species with different body sizes (Burks et al. 2013a). In addition, some males are even more difficult to separate and could not be associated with the females of the same species (Burks et al. 2013a). These issues could be tackled by DNA barcoding (the partial sequencing of the mitochondrial gene cytochrome c oxidase 1, *COI*), which has become an important species identification tool for insects (Hebert et al. 2003a, b). Recently, the Chinese *Oxyscelio* fauna has been reviewed by Mo et al. (2020) and 34 species were recorded, most of which occur in southern China. In this study, we aim to test the morphological species delimitation of *Oxyscelio* species from southern China using DNA barcoding methods.

Materials and methods

Insect sampling and identification

This work is based on specimens of *Oxyscelio* collected by sweep net and mostly Malaise traps (MT) set up in provinces of southern China. Specimens were identified using the keys of Mo et al. (2020). Abbreviations and morphological terms used in text: **A1, A2, ... A12**: antennomere 1, 2, ... 12; **T1, T2, ... T7**: metasomal tergite 1, 2, ... 7; **S1, S2, ... S7**: metasomal sternite 1, 2, ... 7. Morphological terminology otherwise generally follows Burks et al. (2013a) except two terms that are discussed below. The definition of epomial corner used by Burks et al. (2013a) was not clearly stated and refers to where the epomial carina meets the transverse pronotal carina. The corner is weak or strong as a result of the development of the anterior part of the transverse pronotal carina. Therefore, we here describe the development of the anterior part of the transverse pronotal carina instead of the epomial corner. Most *Oxyscelio* species usually have one carina on the middle of the gena, which was termed as ‘middle genal carina’ by Burks et al. (2013a). It seems that Burks et al. (2013a) referred this carina as ‘middle genal carina’ because they erroneously treated the ventral part of the occipital carina as ‘posterior genal carina’. Therefore, we here refer the carina or carinae present on the gena as genal carina or genal carinae.

All the studied specimens are deposited in the Museum of Biology at Sun Yat-sen University, Guangzhou, China (**SYSBM**). Multifocal images were made using a Nikon SMZ25 microscope with a Nikon DS-Ri 2 digital camera system. Images were then post-processed with Adobe Photoshop CS6 Extended.

DNA extraction, amplification, and sequencing

In total, 49 specimens of 22 morphospecies were used for DNA barcoding analysis (see Table 1). Both female and male specimens were selected for each species when such specimens were available. Genomic DNA was extracted from entire specimens using a DNeasy Blood & Tissue Kit (QIAGEN, Inc.), following a nondestructive DNA extraction protocol as described in Taekul et al. (2014). Following DNA extraction, the “barcode” region of the mitochondrial cytochrome oxidase subunit 1 (*COI*) was amplified using the LCO1490/HCO2198 primer pair (Folmer et al. 1994). Polymerase chain reactions (PCRs) were performed using Tks Gflex DNA Polymerase (Takara), and conducted in a T100 Thermal Cycler (Bio-Rad). Thermocycling conditions were: an initial denaturing step at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 50 °C for 30 s, 72 °C for 30 s and an additional extension at 72 °C for 5 min. Amplicons were directly sequenced in both directions with forward and reverse primers on an Applied Biosystems (ABI) 3730XL by Guangzhou Tianyi Huiyuan Gene Technology Co., Ltd. (Guangzhou, China). Chromatograms were assembled with Geneious 11.0.3. All sequences generated from this study are deposited in GenBank (accession numbers see Table 1).

Sequence analysis and molecular species delimitation

All sequences were blasted in BOLD (Barcode of Life Database, http://www.barcodinglife.org/index.php/IDS_OpenIdEngine) and GenBank. Sequences were aligned using MAFFT v7.470 by the G-INS-I strategy (Katoh and Standley 2013). Genetic Kimura-2 parameter (K2P) distances within and between species were calculated in MEGA 7 with pairwise deletion for gaps (Kumar et al. 2016). The aligned sequences were then analyzed using RAXML as implemented in Geneious 11.0.3 under the GTRGAMMA evolutionary model to generate a maximum likelihood (ML) tree. The *COI* sequences of *Probarryconus rufipes* (Kieffer, 1908) (**MF583546**) and *Scelio striatus* Priesner, 1951 (**MF583556**) (Hymenoptera, Scelionidae) were selected as outgroups based on the phylogenetic topologies recovered by Chen et al. (2021).

Results

This study generated 49 *COI* sequences with an average of 660 bp. Voucher specimens of these 49 sequences were subjected to further morphological examination and 22 species were recognized, of which five are described as new. There are only

Table 1. List of sequenced taxa and accession numbers.

Code	Species	Sex	Locality	GenBank accession number
M205	<i>O. amalocarina</i> sp. nov.	female	Hunan, 29°56'N, 110°46'E	MZ539945
M242	<i>O. apheles</i> sp. nov.	female	Yunnan, 21°44.49'N, 100°26.89'E	MZ539946
M182	<i>O. arcus</i>	female	Hainan, 19°4'44.68"N, 109°24'4.74"E	MZ539947
M194	<i>O. arcus</i>	female	Guangdong, 23°10'49.87"N, 113°21'21.33"E	MZ539948
M201	<i>O. arcus</i>	female	Guangdong, 23°9'46.54"N, 112°32'42.9"E	MZ539949
M271	<i>O. arcus</i>	female	Guangxi, 25°21'42.94"N, 110°22'49.9"E	MZ539950
M212	<i>O. brevidentis</i>	male	Yunnan, 21°44.761'N, 100°25.959'E	MZ539951
M213	<i>O. brevidentis</i>	female	Yunnan, 21°44.761'N, 100°25.959'E	MZ539952
M241	<i>O. brevidentis</i>	male	Yunnan, 21°44.498'N, 100°26.889'E	MZ539953
M190	<i>O. convergens</i>	female	Hainan, 19°10'23.28"N, 109°43'40.79"E	MZ539954
M193	<i>O. convergens</i>	male	Hainan, 19°10'23.28"N, 109°43'40.79"E	MZ539955
M195	<i>O. convergens</i>	female	Guangdong, 22°31'53.35"N, 114°36'4.1"E	MZ539956
M197	<i>O. convergens</i>	female	Guangdong, 22°54'26.09"N, 114°13'13.47"E	MZ539957
M198	<i>O. convergens</i>	male	Guangdong, 22°54'26.09"N, 114°13'13.47"E	MZ539958
M203	<i>O. convergens</i>	male	Guangdong, 23°9'45.81"N, 112°32'42.2"E	MZ539959
M204	<i>O. convergens</i>	male	Hainan, 19°4'20.77"N, 109°22'28.2"E	MZ539960
M237	<i>O. convergens</i>	female	Zhejiang, 27°48'45"N, 119°50'28"E	MZ539961
M238	<i>O. convergens</i>	male	Zhejiang, 27°48'45"N, 119°50'28"E	MZ539962
M177	<i>O. crebritas</i>	female	Guangdong, 23°37.287"N, 113°51.267'E	MZ539963
M186	<i>O. crebritas</i>	female	Hainan, 19°10'22.14"N, 109°43'10.34"E	MZ539964
M199	<i>O. crebritas</i>	male	Guangdong, 22°54'26.09"N, 114°13'13.47"E	MZ539965
M222	<i>O. doumao</i>	female	Yunnan, 21°44.745'N, 100°26.07'E	MZ539966
M180	<i>O. excavates</i>	male	Hainan, 18°41'N, 108°49'E	MZ539967
M220	<i>O. granorum</i>	male	Yunnan, 21°44.745'N, 100°26.07'E	MZ539968
M223	<i>O. granorum</i>	male	Yunnan, 21°44.745'N, 100°26.07'E	MZ539969
M226	<i>O. granorum</i>	female	Yunnan, 21°44.912'N, 100°26.647'E	MZ539970
M268	<i>O. intermedietas</i>	female	Yunnan, 21°36'54"N, 101°35'2"E	MZ539971
M269	<i>O. jugi</i>	male	Yunnan, 21°36'54"N, 101°35'2"E	MZ539972
M233	<i>O. kramatos</i>	female	Guangdong, 24°43'N, 114°14'E	MZ539973
M184	<i>O. kiefferi</i>	female	Hainan, 19°4'44.68"N, 109°24'4.74"E	MZ539974
M210	<i>O. labis</i>	female	Yunan, 21°44.761'N, 100°25.959'E	MZ539975
M211	<i>O. labis</i>	male	Yunan, 21°44.761'N, 100°25.959'E	MZ539976
M225	<i>O. labis</i>	male	Yunnan, 21°45.061'N, 100°21.661'E	MZ539977
M185	<i>O. latinubbin</i>	female	Hainan, 19°4'43.32"N, 109°23'38.74"E	MZ539978
M206	<i>O. latheticus</i> sp. nov.	female	Hunan, 29°56'N, 110°46'E	MZ539979
M232	<i>O. latheticus</i> sp. nov.	female	Guangdong, 24°43'N, 114°14'E	MZ539980
M183	<i>O. nubbin</i>	female	Hainan, 19°4'44.68"N, 109°24'4.74"E	MZ539981
M187	<i>O. nubbin</i>	female	Hainan, 19°10'22.14"N, 109°43'10.34"E	MZ539982
M188	<i>O. nubbin</i>	male	Hainan, 19°10'22.14"N, 109°43'10.34"E	MZ539983
M192	<i>O. nubbin</i>	female	Hainan, 19°30'56.91"N, 109°29'29.28"E	MZ539984
M179	<i>O. nullicarina</i>	male	Hainan, 18°41'N, 108°49'E	MZ539985
M231	<i>O. nullicarina</i>	female	Yunnan, 22°52'53.09"N, 104°5'50.98"E	MZ539986
M216	<i>O. ogive</i>	female	Yunnan, 21°44.746'N, 100°26'E	MZ539987
M219	<i>O. ogive</i>	female	Yunnan, 21°44.745'N, 100°26.07'E	MZ539988
M239	<i>O. stenosis</i> sp. nov.	female	Yunnan, 23°14'11.18"N, 104°6'3.44"E	MZ539989
M215	<i>O. striae</i> sp. nov.	female	Yunnan, 21°44.746'N, 100°26'E	MZ539990
M275	<i>O. striae</i> sp. nov.	female	Yunnan, 21°44.746'N, 100°26'E	MZ539991
M278	<i>O. striae</i> sp. nov.	female	Yunnan, 21°44.746'N, 100°26'E	MZ539992
M229	<i>O. striarum</i>	female	Yunnan, 21°91.613'N, 101°27.118'E	MZ539993

two *COI* sequences of *Oxyscelio* spp. in the BOLD and GenBank databases and the blast results of the sequences generated in this study returned no matching sequences. Genetic distances of the 49 sequences are in Suppl. material 1: Table S1.

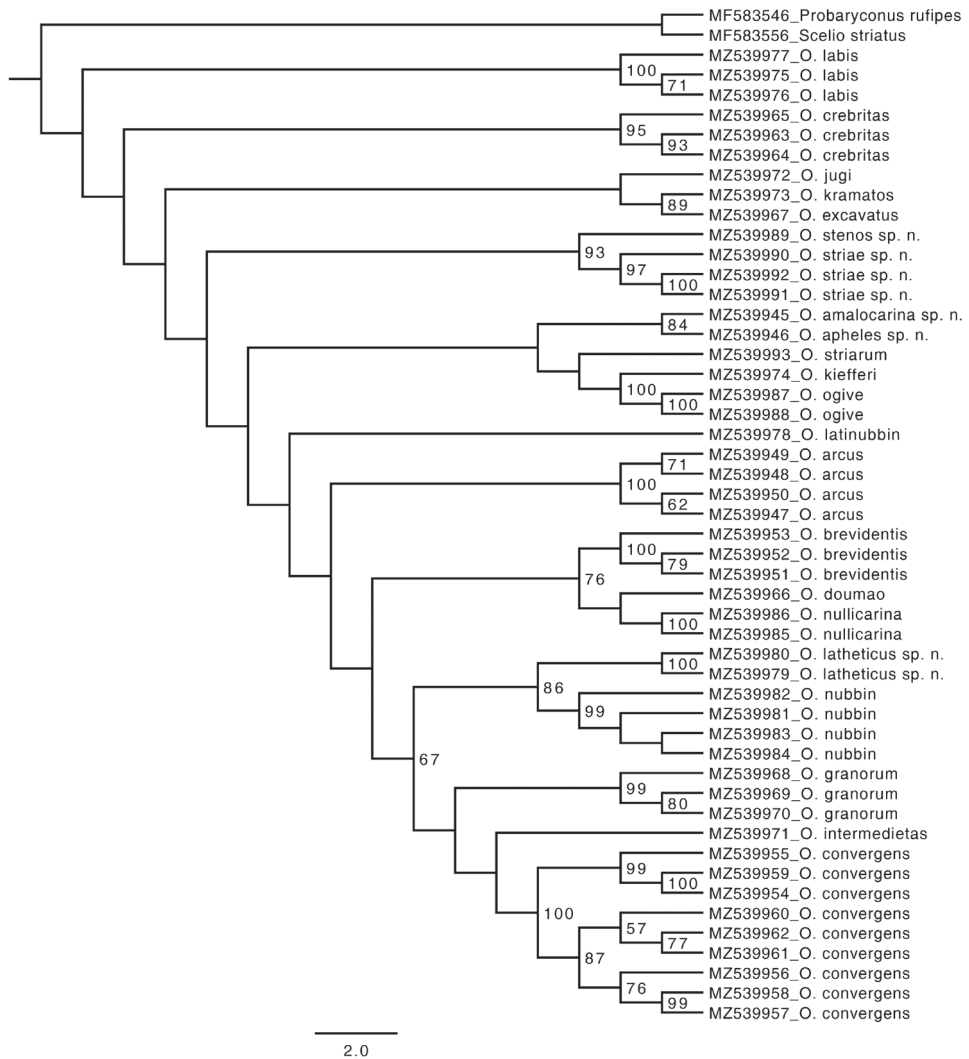


Figure 1. Maximum likelihood tree demonstrating the clustering of *Oxyscelio* COI barcodes. Bootstraps values of 50 and above are indicated.

Intraspecific distances of the COI sequences generally are less than 3%, with two exceptions: *O. crebritas* and *O. convergens*. *Oxyscelio crebritas* has two haplogroups and the distance between the two haplogroups is 4.3%. *Oxyscelio convergens* also has two haplogroups and the distance between the two haplogroups is 3.9%. However, the two haplogroups of these two species are both clustered together on the ML tree (Figure 1), respectively. Interspecific distances range between 7.1% and 19%. Overall, the morphology-based delimitations of species are generally highly supported, as shown in Figure 1, indicating that DNA barcoding is well suited to help resolve species limits.

Species treatment

Oxyscelio amalocarina Mo & Chen, sp. nov.

<http://zoobank.org/B1EA6947-3B08-41B1-BDD5-3E21D7BB8A44>

Figure 2

Description. Female. Body length 3.65 mm ($n = 1$).

Radicle color: same color as scape. Scape color: yellowish. A4: longer than broad. A5: longer than broad. Antennal club in female: formed, segments compact.

Interantennal process: not elongate. Median longitudinal elevation in frontal depression: absent. Frontal depression: flat. Frontal depression sculpture: with 3–5 complete transverse carinae below submedian carina. Submedian carina: rather weak, only upper portion developed. Submedian carina medially: without peak. Concavity across dorsal part of frontal depression: absent. Depression extending ventrally from median ocellus: absent. Upper frons: not hood-like. Malar area near antennal foramen: without carina or expansion. Facial striae: present. Smooth strip along posterior side of malar sulcus: present, broad throughout its length. Genal carina: absent. Major sculpture of gena anteriorly: punctate. Major sculpture of gena posteriorly: rugose; punctate. Microsculpture of gena anteroventrally: absent. Microsculpture of gena posteroventrally: absent. Median carina extending posteriorly from hyperoccipital carina: absent. Hyperoccipital carina: indicated by rugae. Lateral connection between hyperoccipital and occipital carinae: absent. Area between vertex and occipital carina: foveate. Occipital carina medially: absent. Lateral corners of occipital carina: sharp and protruding.

Lateral pronotal area: without bulge projecting towards anterior pit. Transverse pronotal carina: absent anteriorly. Netrion surface anteriorly: not inflected. Mesoscutum anteriorly: steep. Mesoscutal median carina: present and complete. Longitudinal carina between median carina and notauli: absent. Major sculpture of medial mesoscutum anteriorly: foveate; rugose. Major sculpture of medial mesoscutum posteriorly: foveate. Microsculpture of medial mesoscutum anteriorly: granulate. Microsculpture of medial mesoscutum posteriorly: absent. Major sculpture of mesoscutellum: foveate. Microsculpture of mesoscutellum medially: absent. Microsculpture of mesoscutellum laterally: absent. Mesoscutellar apex: straight. Setae along anterior limit of femoral depression: arising from rows of foveae. Number of carinae crossing speculum above femoral depression: 3. Number of carinae crossing femoral depression: more than 5. Mesepimeral sulcus pits: more than 5. Metascutellum dorsally: concave. Metascutellar sculpture dorsally: areolate. Median carina of metascutellum: absent. Metascutellar setae: with many dorsal setae. Metascutellar apex: weakly emarginate. Metapleuron above ventral metapleural area: crossed by carinae. Metasomal depression setae: not visible. Lateral propodeal carinae anteromedially: strongly diverging. Anterior areoles of metasomal depression: not visible. Anterior longitudinal carinae in metasomal depression: not visible. Lateral propodeal areas: separated medially. Postmarginal vein: absent. Fore wing apex: reaching apex of T5.

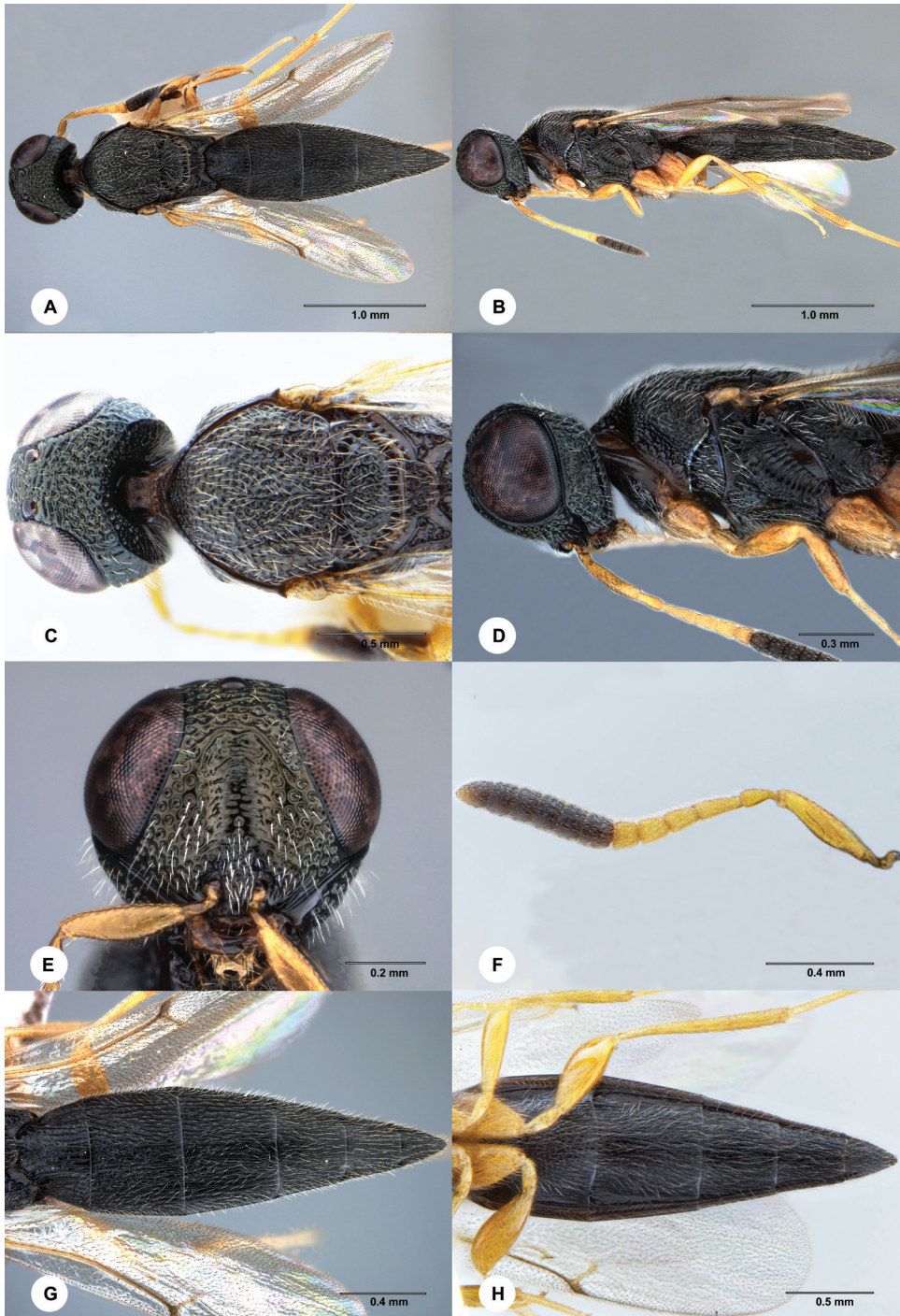


Figure 2. *Oxyscelio amalocarina* Mo & Chen, sp. nov., holotype, female (SCAU 3049046) **A** dorsal habitus **B** lateral habitus **C** head and mesosoma, dorsal view **D** head and mesosoma, lateral view **E** head, anterior view **F** antenna **G** metasoma, dorsal view **H** metasoma, ventral view.

T1 midlobe: obscured by other raised sculpture. T1: with anterior bulge. T2: with straight longitudinal striae. T6: longer than broad. Metasomal apex: rounded. Major sculpture of T6: punctate. Microsculpture of T6: absent. S2: with longitudinal rugae. S5: with longitudinal striae. S6: punctate.

Male. Unknown.

Diagnosis. Female: A4 and A5 longer than broad. Frontal depression flat. Submedian carina of frons weak, only upper portion developed. Gena without middle carina. Occipital carina absent medially. Metascutellum areolate, with many dorsal setae. T1 with anterior bulge. The above diagnostic characters make *O. amalocarina* unique and can be easily separated from other species of *Oxyscelio*.

Etymology. The name *amalocarina* refers to the weak submedian carina of this species. The epithet is used as a noun in apposition.

Material examined. *Holotype*, female: CHINA: Hunan, Mt. Hupingshan, 29°56'N, 110°46'E, 9.VIII.2009, Ya-li Tang, SCAU 3049046 (deposited in SYSBM).

Distribution. China (Hunan).

The key to females of the Chinese *Oxyscelio* published by Mo et al. (2020) could be updated to accommodate *O. amalocarina* by replacing couplet 6 as follows:

- | | |
|----|--|
| 6 | Mesosoma and metasoma black (Fig. 2A); mesoscutum foveate posteriorly (Fig. 2C) 6' |
| – | Mesosoma and metasoma yellowish; mesoscutum transversely rugose posteriorly <i>Oxyscelio nullicarina</i> Mo & Chen |
| 6' | Upper frons not hood-like (Fig. 2E); T1 with anterior bulge (Fig. 2G) <i>Oxyscelio amalocarina</i> Mo & Chen, sp. nov. |
| – | Upper frons hood-like; T1 without anterior bulge <i>Oxyscelio doumao</i> Burks |

***Oxyscelio apheles* Mo & Chen, sp. nov.**

<http://zoobank.org/E44FFEC7-ABC5-46D1-B4A7-C4F9B264BDAC>

Figure 3

Description. Female. Body length 3.08 mm (n = 1).

Radicle color: same color as scape. Scape color: dark brown. A4: as long as broad. A5: broader than long. Antennal club in female: formed, segments compact.

Interantennal process: not elongate. Median longitudinal elevation in frontal depression: absent. Frontal depression: concave. Frontal depression sculpture: with 3 broadly interrupted transverse carinae below submedian carina. Submedian carina: strong, formed by a sharp raised carina. Submedian carina medially: with sharp peak. Concavity across dorsal part of frontal depression: absent. Depression extending ventrally from median ocellus: absent. Upper frons: not hood-like. Malar area near antennal foramen: without carina or expansion. Facial striae: present. Smooth strip along

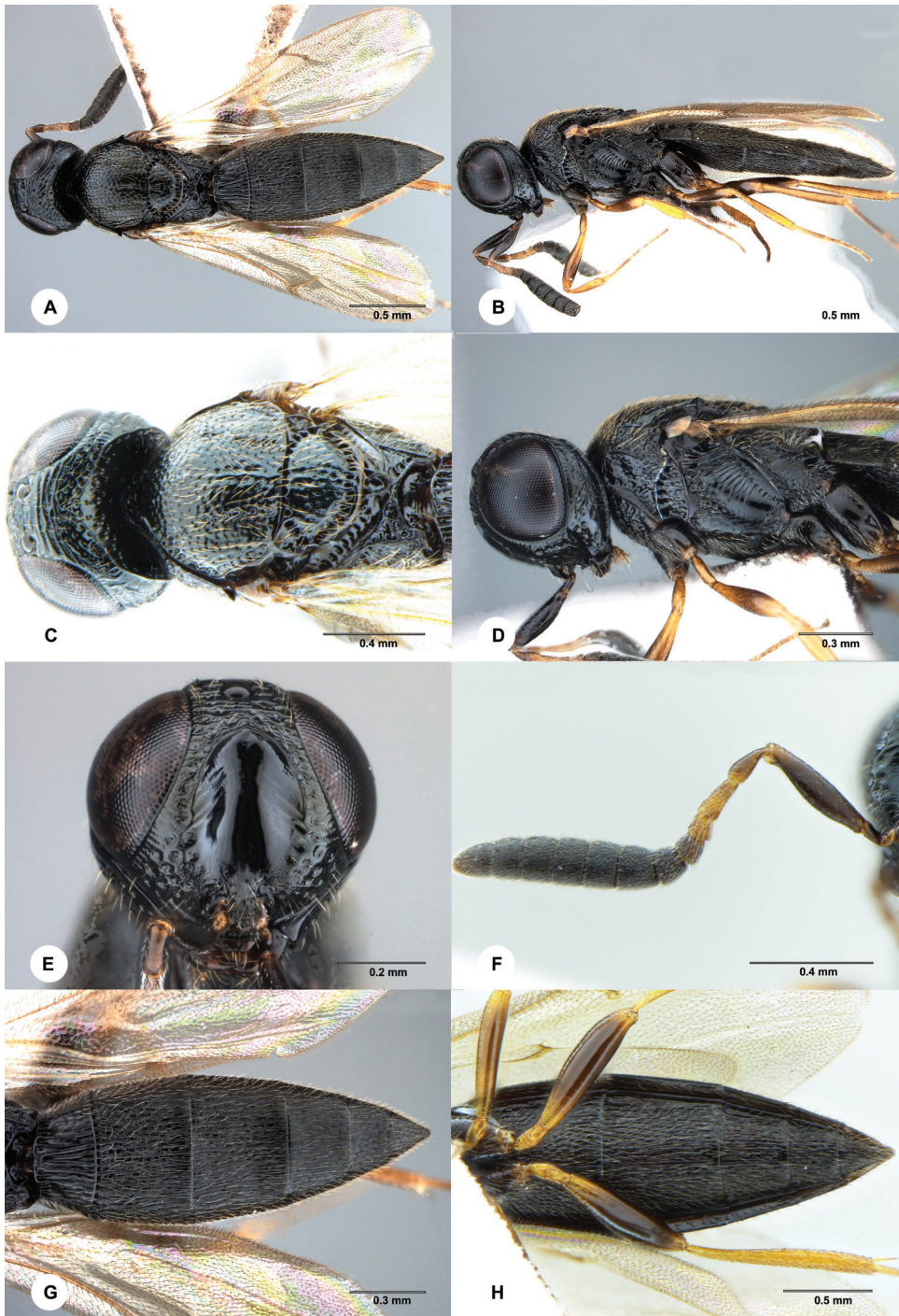


Figure 3. *Oxyscelio apheles* Mo & Chen, sp. nov., holotype, female (SCAU 3049046) **A** dorsal habitus **B** lateral habitus **C** head and mesosoma, dorsal view **D** head and mesosoma, lateral view **E** head, anterior view **F** Antenna **G** metasoma, dorsal view **H** metasoma, ventral view.

posterior side of malar sulcus: absent or not consistently broad. Genal carina: present. Direction of genal carina dorsally: parallel to eye margin. Major sculpture of gena anteriorly: smooth with scattered punctures. Major sculpture of gena posteriorly: rugulose with scattered punctures. Microsculpture of gena anteroventrally: absent. Microsculpture of gena posteroventrally: absent. Median carina extending posteriorly from hyperoccipital carina: absent. Hyperoccipital carina: indicated by rugae. Lateral connection between hyperoccipital and occipital carinae: absent. Area between vertex and occipital carina: foveate. Occipital carina medially: uniformly rounded. Lateral corners of occipital carina: protruding.

Lateral pronotal area: without bulge projecting towards anterior pit. Transverse pronotal carina: present anteriorly and forming a corner with epomial carina. Notrion surface anteriorly: not inflected. Mesoscutum anteriorly: steep. Mesoscutal median carina: present and complete. Longitudinal carina between median carina and notauli: absent. Major sculpture of medial mesoscutum anteriorly: punctate. Major sculpture of medial mesoscutum posteriorly: punctate. Microsculpture of medial mesoscutum anteriorly: granulate. Microsculpture of medial mesoscutum posteriorly: absent. Major sculpture of mesoscutellum: punctate. Microsculpture of mesoscutellum medially: absent. Microsculpture of mesoscutellum laterally: absent. Mesoscutellar apex: convex or straight. Setae along anterior limit of femoral depression: arising from rows of foveae. Number of carinae crossing speculum above femoral depression: 3. Number of carinae crossing femoral depression: more than 5. Mesepimeral sulcus pits: more than 5. Metascutellum dorsally: concave. Metascutellar sculpture dorsally: smooth. Median carina of metascutellum: absent. Metascutellar setae: absent. Metascutellar apex: straight. Metapleuron above ventral metapleural area: crossed by carinae. Metasomal depression setae: absent. Lateral propodeal carinae anteromedially: weakly diverging. Anterior areoles of metasomal depression: absent. Anterior longitudinal carinae in metasomal depression: absent. Lateral propodeal areas: separated medially. Postmarginal vein: absent. Fore wing apex: reaching beyond T6.

T1 midlobe: with 6 longitudinal carinae. T1: without anterior bulge. T2: with straight longitudinal striae or rugae. T6: broader than long. Metasomal apex: rounded. Major sculpture of T6: punctate. Microsculpture of T6: absent. S2: with longitudinal rugae. S5: with longitudinal striae. S6: punctate rugose.

Male. Unknown.

Diagnosis. Female: A4 as long as broad, A5 broader than long. Frons without elevation between antennal foramen and eye. Upper frons not hood-like. Hyperoccipital carina indicated by rugae. Medial mesoscutum punctate posteriorly. Metascutellum without dorsal setae, smooth. T1 with 6 longitudinal carinae. *Oxyscelio apheles* is similar to *O. vittae* Burks but can be separated from the latter by the following characters: radicle, scape and coxae blackish, metapleuron above ventral metapleural area crossed by carinae.

Etymology. The name *apheles* refers to the smooth metascutellum of this species. The epithet is used as a noun in apposition.

Material examined. *Holotype*, female: CHINA: Yunnan, Xishuangbanna, Menghai, Bulangshan Village, 1659 m, Area B1, forest, 21°44.498'N, 100°26.889'E, 28.VI–19.VII.2019, Li Ma, SCAU 3049083 (deposited in SYSBM).

Distribution. China (Yunnan).

The key to females of the Chinese *Oxyscelio* published by Mo et al. (2020) could be updated to accommodate *O. apheles* by replacing couplet 23 as follows:

- 23 Genal carina strong, with two strong carinae below; postmarginal vein present..... *Oxyscelio labis* Burks
- Genal carina weakly indicated, without carinae below; postmarginal vein absent..... 23'
- 23' Submedian carina with sharp peak medially (Fig. 3E); radicle and scape dark brown (Fig. 3F)..... *Oxyscelio apheles* Mo & Chen, sp. nov.
- Submedian carina without sharp peak medially; radicle and scape yellowish .
..... *Oxyscelio naraws* Burks

***Oxyscelio latheticus* Mo & Chen, sp. nov.**

<http://zoobank.org/681C9A5A-7E92-42EA-B71B-9A9B5096D693>

Figure 4

Description. Female. Body length 2.9–2.93 mm (n = 2).

Radicle color: same color as scape. Scape color: yellowish. A4: broader than long. A5: broader than long. Antennal club in female: formed, segments compact.

Interantennal process: not elongate. Median longitudinal elevation in frontal depression: absent. Frontal depression: concave. Frontal depression sculpture: with 3 broadly interrupted transverse carinae below submedian carina. Submedian carina: strong, formed by a sharp raised carina. Submedian carina medially: without peak. Concavity across dorsal part of frontal depression: absent. Depression extending ventrally from median ocellus: absent. Upper frons: hood-like. Malar area near antennal foramen: without carina or expansion. Facial striae: present. Smooth strip along posterior side of malar sulcus: absent or not consistently broad. Genal carina: present. Direction of genal carina dorsally: parallel to eye margin. Major sculpture of gena anteriorly: rugose; foveate. Major sculpture of gena posteriorly: rugose; foveate. Microsculpture of gena anteroventrally: absent. Microsculpture of gena posteroventrally: absent. Median carina extending posteriorly from hyperoccipital carina: present. Hyperoccipital carina: complete, extending ventrally around outer orbit of compound eye. Lateral connection between hyperoccipital and occipital carinae: absent. Area between vertex and occipital carina: irregularly rugose. Occipital carina medially: uniformly rounded. Lateral corners of occipital carina: not protruding.

Lateral pronotal area: without bulge projecting towards anterior pit. Transverse pronotal carina: present anteriorly and forming a corner with epomial carina. Netrion

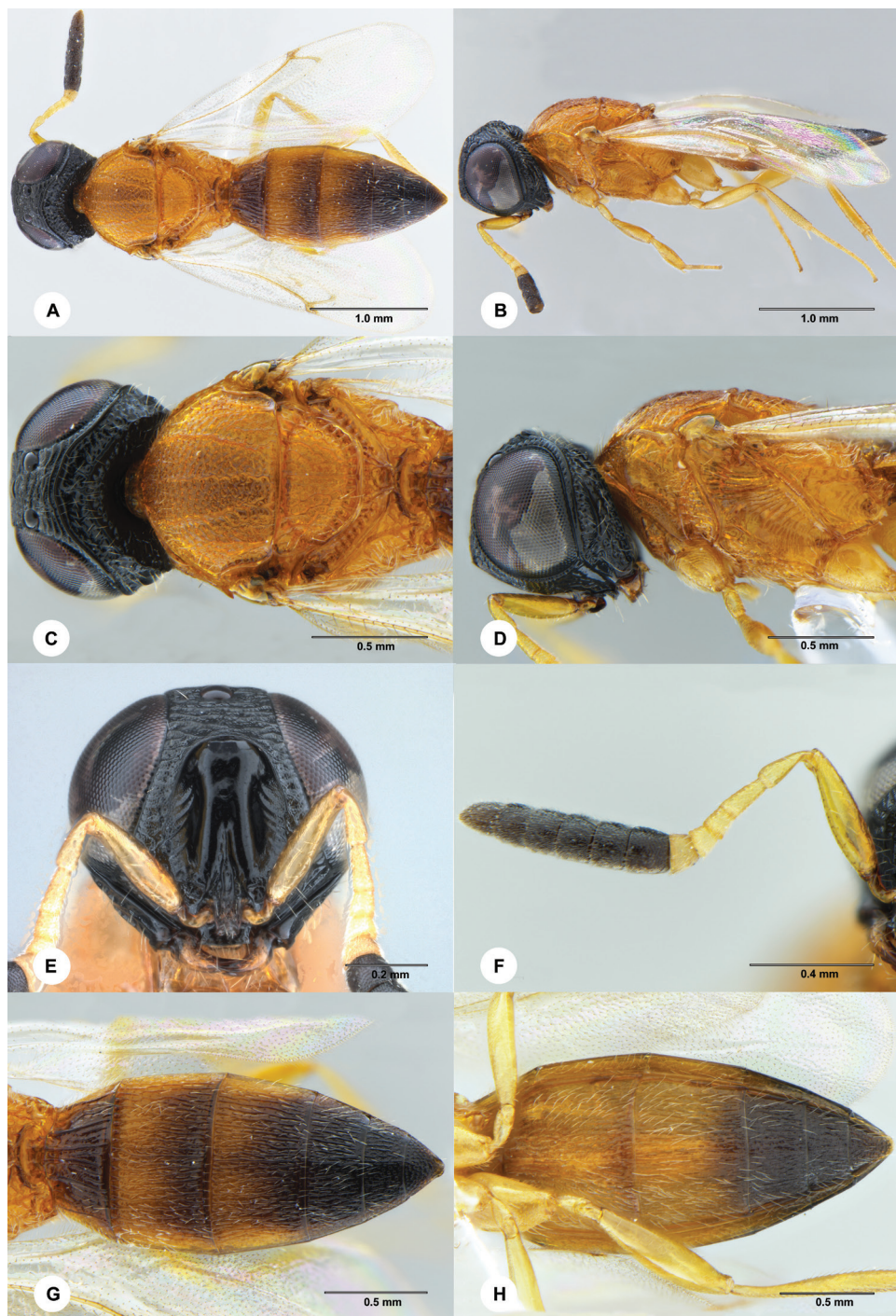


Figure 4. *Oxyscelio latheticus* Mo & Chen, sp. nov., holotype, female (SCAU 3049073) **A** dorsal habitus **B** lateral habitus **C** head and mesosoma, dorsal view **D** head and mesosoma, lateral view **E** head, anterior view **F** antenna **G** metasoma, dorsal view **H** metasoma, ventral view.

surface anteriorly: not inflected. Mesoscutum anteriorly: steep. Mesoscutal median carina: present and complete. Longitudinal carina between median carina and notauli: absent. Major sculpture of medial mesoscutum anteriorly: foveate; rugose. Major sculpture of medial mesoscutum posteriorly: foveate. Microsculpture of medial mesoscutum anteriorly: granulate. Microsculpture of medial mesoscutum posteriorly: granulate. Major sculpture of mesoscutellum: foveate; longitudinally rugose. Microsculpture of mesoscutellum medially: granulate. Microsculpture of mesoscutellum laterally: granulate. Mesoscutellar apex: convex or straight. Setae along anterior limit of femoral depression: arising from rows of foveae. Number of carinae crossing speculum above femoral depression: 4. Number of carinae crossing femoral depression: more than 5. Mesepimeral sulcus pits: more than 5. Metascutellum dorsally: concave. Metascutellar sculpture dorsally: foveate. Median carina of metascutellum: absent. Metascutellar setae: absent. Metascutellar apex: straight. Metapleuron above ventral metapleural area: crossed by carinae. Metasomal depression setae: absent. Lateral propodeal carinae anteromedially: strongly diverging. Anterior areoles of metasomal depression: absent. Anterior longitudinal carinae in metasomal depression: absent. Lateral propodeal areas: separated medially. Postmarginal vein: present. Fore wing apex: reaching beyond T6.

T1 midlobe: with 4 longitudinal carinae. T1: without anterior bulge. T2: with straight longitudinal striae. T6: broader than long. Metasomal apex: rounded. Major sculpture of T6: punctate. Microsculpture of T6: granulate. S2: with longitudinal rugae. S5: with longitudinal rugae. S6: punctate rugose.

Male. Unknown.

Diagnosis. Female: A4 and A5 broader than long. Frons without elevation between antennal foramen and eye. Hyperoccipital carina present, extending ventrally around outer orbit of compound eye. Medial mesoscutum foveate. Metascutellum without dorsal setae. T1 with 4 longitudinal carinae. *Oxyscelio latheticus* is extremely similar to *O. convergens* but can be distinguished from the latter by the following characters: A4 and A5 broader than long in *O. latheticus* (longer than broad in *O. convergens*); metascutellar apex straight and the lateral carina of metascutellum blunt, while in *O. convergens*, metascutellar apex emarginated and the lateral carina of metascutellum forming an acute tooth.

Etymology. The name *latheticus* refers to the fact that this species is extremely similar to *O. convergens* and could be overlooked. The epithet is used as a noun in apposition.

Material examined. *Holotype*, female: CHINA: Guangdong, Mt. Chebaling, 24°43'N, 114°14'E, 22–28.II.2008, Zai-fu Xu, SCAU 3049073 (deposited in SYS-BM). *Paratype*: Hunan, Mt. Hupingshan, 29°56'N 110°46'E, 9.VII.2009, Ya-li Tang, SCAU 3049047 (SYSBM)

Distribution. China (Hunan, Guangdong).

The key to females of the Chinese *Oxyscelio* published by Mo et al. (2020) could be updated to accommodate *O. latheticus* by replacing couplet 10 as follows:

- | | | |
|----|---|-----|
| 10 | Mesosoma and metasoma yellowish..... | 10' |
| – | Mesosoma and metasoma entirely black..... | 11 |

- 10' A5 broader than long (Fig. 4F); metascutellar apex straight and the lateral carina of metascutellum blunt (Fig. 4C).....
 *Oxyscelio latheticus* Mo & Chen, sp. nov.
 – A5 longer than broad; metascutellar apex emarginated and the lateral carina of metascutellum forming an acute tooth..... *Oxyscelio convergens* Burks

***Oxyscelio stenos* Mo & Chen, sp. nov.**

<http://zoobank.org/0CA2EB89-BE13-44FB-95D8-C9F5036BBA89>

Figure 5

Description. Female. Body length 2.83 mm (n = 1).

Radicle color: darker than scape. Scape color: yellowish. A4: broader than long. A5: broader than long. Antennal club in female: formed, segments compact.

Interantennal process: not elongate. Median longitudinal elevation in frontal depression: present. Frontal depression: concave. Frontal depression sculpture: with 3 broadly interrupted transverse carinae below submedian carina. Submedian carina: strong, formed by a sharp raised carina. Submedian carina medially: with sharp peak. Concavity across dorsal part of frontal depression: absent. Depression extending ventrally from median ocellus: absent. Upper frons: not hood-like. Malar area near antennal foramen: without carina or expansion. Facial striae: present. Smooth strip along posterior side of malar sulcus: absent or not consistently broad. Genal carina: present. Direction of genal carina dorsally: parallel to eye margin. Major sculpture of gena anteriorly: rugose; foveate. Major sculpture of gena posteriorly: rugose; foveate. Microsculpture of gena anteroventrally: absent. Microsculpture of gena posteroventrally: granulate. Median carina extending posteriorly from hyperoccipital carina: present. Hyperoccipital carina: complete, continuous with anterior genal carina. Lateral connection between hyperoccipital and occipital carinae: present. Area between vertex and occipital carina: irregularly rugose. Occipital carina medially: convex, with a sharp median peak. Lateral corners of occipital carina: sharp and protruding.

Lateral pronotal area: without bulge projecting towards anterior pit. Transverse pronotal carina: present anteriorly and forming a corner with epomial carina. Notrion surface anteriorly: not inflected. Mesoscutum anteriorly: steep. Mesoscutal median carina: present and complete. Longitudinal carina between median carina and notauli: absent. Major sculpture of medial mesoscutum anteriorly: punctate; rugose. Major sculpture of medial mesoscutum posteriorly: punctate. Microsculpture of medial mesoscutum anteriorly: granulate. Microsculpture of medial mesoscutum posteriorly: absent. Major sculpture of mesoscutellum: punctate; longitudinally rugose. Microsculpture of mesoscutellum medially: absent. Microsculpture of mesoscutellum laterally: absent. Mesoscutellar apex: straight. Setae along anterior limit of femoral depression: arising from rows of foveae. Number of carinae crossing speculum above femoral depression: 2. Number of carinae crossing femoral depression: more than 5. Mesepimeral sulcus pits: more than 5. Metascutellum dorsally: concave. Metascutellar

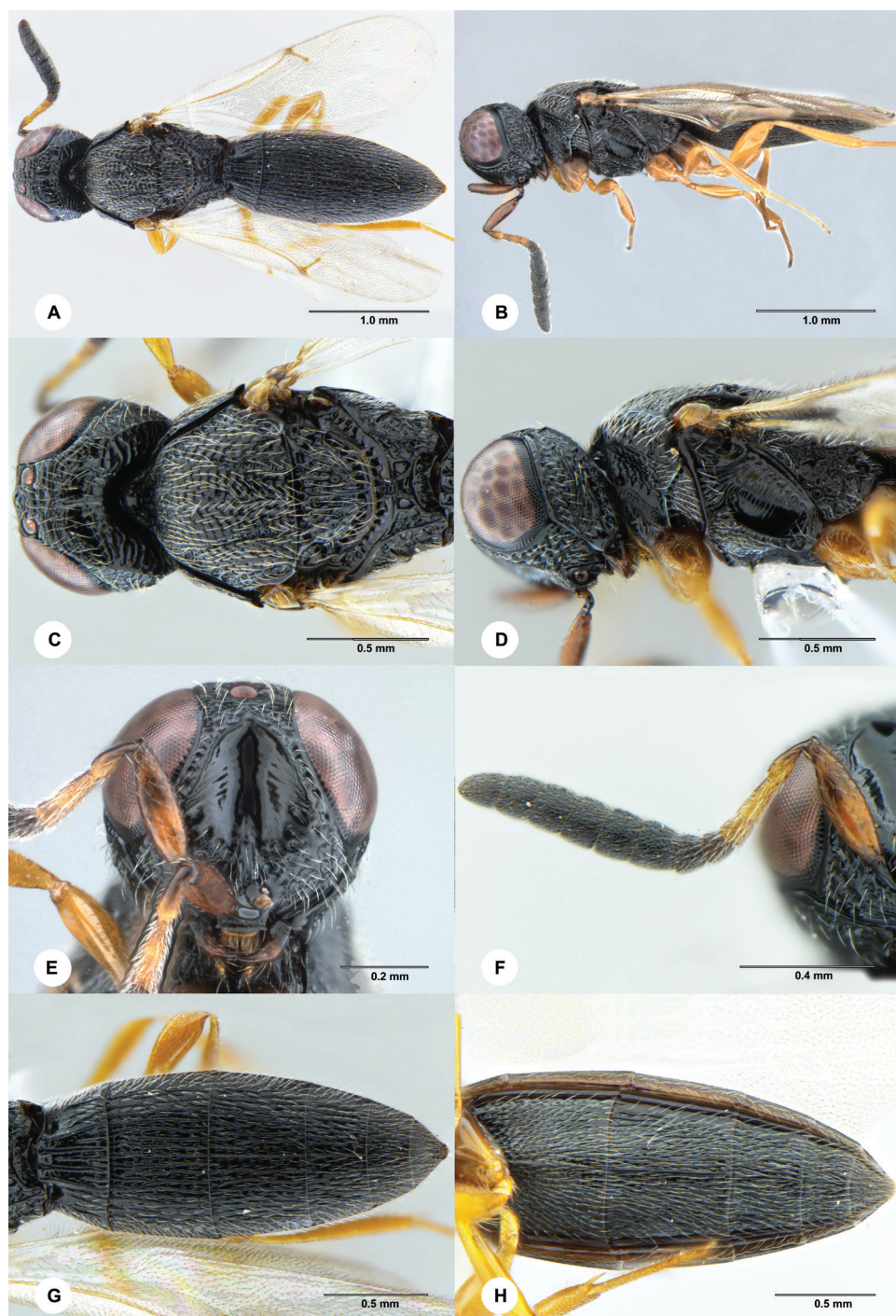


Figure 5. *Oxyscelio stenos* Mo & Chen, sp. nov., holotype, female (SCAU 3049080) **A** dorsal habitus **B** lateral habitus **C** head and mesosoma, dorsal view **D** head and mesosoma, lateral view **E** head, anterior view **F** antenna **G** metasoma, dorsal view **H** metasoma, ventral view.

sculpture dorsally: foveate. Median carina of metascutellum: absent. Metascutellar setae: absent. Metascutellar apex: straight. Metapleuron above ventral metapleural area: foveate. Metasomal depression setae: absent. Lateral propodeal carinae anteromedially: strongly diverging. Anterior areoles of metasomal depression: present. Anterior longitudinal carinae in metasomal depression: present. Lateral propodeal areas: separated medially. Postmarginal vein: absent. Fore wing apex: reaching beyond apex of T6.

T1 midlobe: with 4 longitudinal carinae. T1: without anterior bulge. T2: with straight longitudinal striae. T6: broader than long. Metasomal apex: rounded. Major sculpture of T6: punctate. Microsculpture of T6: absent. S2: with longitudinal rugae. S5: with longitudinal striae. S6: punctate.

Male. Unknown.

Diagnosis. Female: A4 and A5 broader than long. Frontal depression with median longitudinal elevation. Frons without elevation between antennal foramen and eye. Hyperoccipital carina present, extending ventrally around outer orbit of compound eye. Medial mesoscutum punctate. Metascutellum without dorsal setae. T1 with 4 longitudinal carinae. *Oxyscelio stenosis* is similar to *O. ogive* and *O. striae* but can be separated from the latter two species by the narrow metascutellum and the median longitudinal elevation on frontal depression (see the updated key below).

Etymology. The name *stenosis* refers to the narrow metascutellum of this species. The epithet is used as a noun in apposition.

Material examined. *Holotype*, female: CHINA: Yunnan, Wenshan City, Pingba Town, 1793 m, 23.23644°N, 104.100955°E, 31.VIII–22.IX.2009, Malaise trap, SCAU 3049080 (deposited in SYSBM).

Distribution. China (Yunnan).

Oxyscelio striae Mo & Chen, sp. nov.

<http://zoobank.org/D27BEA28-9CBF-49BD-8C06-1D80A5E28FFE>

Figures 6

Description. Female. Body length 2.86–3.0 mm (n = 3).

Radicle color: darker than scape. Scape color: yellowish. A4: broader than long. A5: broader than long. Antennal club in female: formed, segments compact.

Interantennal process: not elongate. Median longitudinal elevation in frontal depression: absent. Frontal depression: concave. Frontal depression sculpture: with 3 broadly interrupted transverse carinae below submedian carina. Submedian carina: strong, formed by a sharp raised carina. Submedian carina medially: with sharp peak. Concavity across dorsal part of frontal depression: absent. Depression extending ventrally from median ocellus: absent. Upper frons: not hood-like. Malar area near antennal foramen: without carina or expansion. Facial striae: present. Smooth strip along posterior side of malar sulcus: absent. Genal carina: present. Direction of genal carina dorsally: parallel to eye margin. Major sculpture of gena anteriorly: foveate above genal carina, striate behind genal carina. Major sculpture of gena posteriorly: rugose.

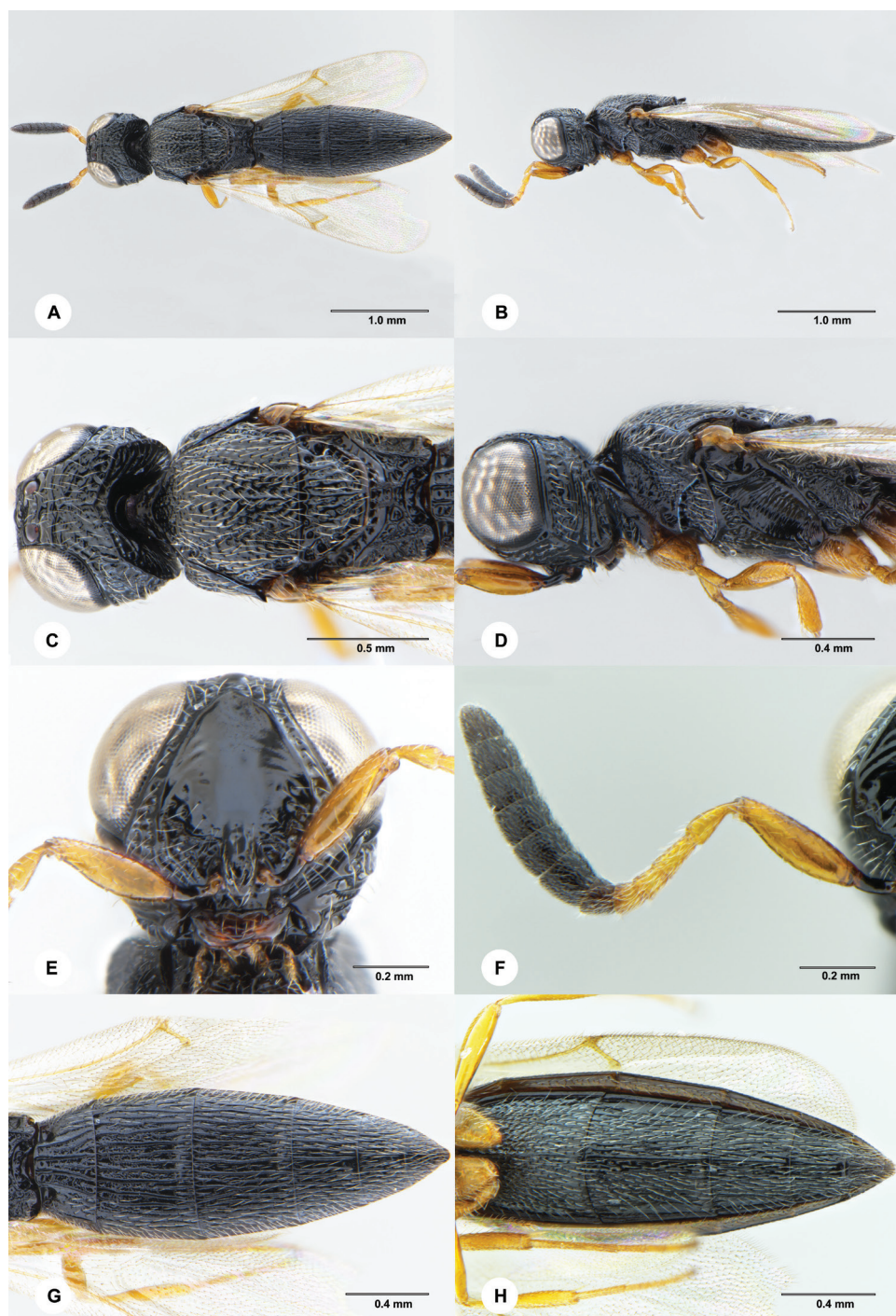


Figure 6. *Oxyscelio striae* Mo & Chen, sp. nov., holotype, female (SCAU 3048667) **A** dorsal habitus **B** lateral habitus **C** head and mesosoma, dorsal view **D** head and mesosoma, lateral view **E** head, anterior view **F** antenna **G** metasoma, dorsal view **H** metasoma, ventral view.

Microsculpture of gena anteroventrally: absent. Microsculpture of gena posteroventrally: granulate. Median carina extending posteriorly from hyperoccipital carina: present. Hyperoccipital carina: complete, extending ventrally around outer orbit of compound eye. Lateral connection between hyperoccipital and occipital carinae: present. Area between vertex and occipital carina: rugose; foveate. Occipital carina medially: convex, with a sharp median peak. Lateral corners of occipital carina: sharp and protruding.

Lateral pronotal area: without bulge projecting towards anterior pit. Transverse pronotal carina: present anteriorly and forming a corner with epomial carina. Netrion surface anteriorly: not inflected. Mesoscutum anteriorly: steep. Mesoscutal median carina: present and complete. Longitudinal carina between median carina and notauli: absent. Major sculpture of medial mesoscutum anteriorly: punctate; rugose. Major sculpture of medial mesoscutum posteriorly: punctate. Microsculpture of medial mesoscutum anteriorly: granulate. Microsculpture of medial mesoscutum posteriorly: granulate. Major sculpture of mesoscutellum: foveate; longitudinally rugose. Microsculpture of mesoscutellum medially: absent. Microsculpture of mesoscutellum laterally: absent. Mesoscutellar apex: straight. Setae along anterior limit of femoral depression: arising from rows of foveae. Number of carinae crossing speculum above femoral depression: 3. Number of carinae crossing femoral depression: more than 5. Mesepimeral sulcus pits: more than 5. Metascutellum dorsally: concave. Metascutellar sculpture dorsally: rugose. Median carina of metascutellum: absent. Metascutellar setae: absent. Metascutellar apex: convex. Metapleuron above ventral metapleural area: rugose. Metasomal depression setae: absent. Lateral propodeal carinae anteromedially: strongly diverging. Anterior areoles of metasomal depression: present. Anterior longitudinal carinae in metasomal depression: absent. Lateral propodeal areas: separated medially. Postmarginal vein: absent. Fore wing apex: reaching beyond T5.

T1 midlobe: with 4 longitudinal carinae. T1: without anterior bulge. T2: with straight longitudinal striae. T6: broader than long. Metasomal apex: rounded. Major sculpture of T6: punctate. Microsculpture of T6: absent. S2: with longitudinal rugae. S5: with longitudinal striae. S6: punctate.

Male. Unknown.

Diagnosis. Female: A4 and A5 broader than long. Frons without elevation between antennal foramen and eye. Gena with 3 striae below genal carina. Hyperoccipital carina present, extending ventrally around outer orbit of compound eye. Medial mesoscutum punctate. Metascutellum without dorsal setae. T1 with 4 longitudinal carinae. *Oxyscelio striae* is similar to *O. ogive* but can be separated from the latter by the darker radicle (yellowish in *O. ogive*), rugose metascutellum (smooth in *O. ogive*).

Etymology. The name *striae* refers to the striate gena of this species. The epithet is used as a noun in apposition.

Material examined. *Holotype*, female: **CHINA:** Yunnan, Xishuangbanna, Menghai, Bulangshan Village, 1610 m, Area D, grass, 21°44.761'N, 100°26'E, 28.V–28.VI.2019, Li Ma, Malaise trap, SCAU 3048667 (deposited in SYSBM). Paratypes: (2 females) **CHINA:** 1 female, same data as holotype, SCAU 3048664 (SYSBM); 1 female, same data as holotype, except 20.VI–20.VII.2018, SCAU 3049056 (SYSBM).

Distribution. China (Yunnan).

The key to females of the Chinese *Oxyscelio* published by Mo et al. (2020) could be updated to accommodate *O. stenos* and *O. striae* by replacing couplet 22 as follows:

- 22 Mesoscutellum with a strong median carina.....22'
- Mesoscutellum without a strong median carina23
- 22' Frontal depression with median longitudinal elevation (Fig. 5E); genal carina without adjacent striae (Fig. 5D) ***Oxyscelio stenos* Mo & Chen, sp. nov.**
- Frontal depression without median longitudinal elevation; genal carina with adjacent striae22''
- 22'' Radicle yellowish; metascutellum smooth dorsally ***Oxyscelio ogive* Burks**
- Radicle dark brown (Fig. 6F); metascutellum rugose dorsally (Fig. 6C)
..... ***Oxyscelio striae* Mo & Chen, sp. nov.**

Discussion

In the last few years, DNA barcode-based approaches have become increasingly popular for the identification of Scelionidae, especially for economically important genera such as *Trissolcus* Ashmead, *Telenomus* Haliday and *Gryon* Haliday (Liao et al. 2019; Talamas et al. 2019; Chen et al. 2020). These approaches not only provide evidence of species boundaries that supplement morphological identifications, but also become invaluable means of confirmation in some cases (Talamas et al. 2019). In this study, we provide *COI* sequences for 22 species of *Oxyscelio*, comprising more than half of all the species of the genus known from China. Our results indicate that the use of DNA barcoding for the identification of *Oxyscelio* species is promising. For example, Burks et al. (2013a) noted that *O. intermedietas* might be a small-bodied relative of *O. granorum*, but both species were treated as new species based on several subtle differences. Our molecular data support the treatment of *O. intermedietas* and *O. granorum* as distinct species (Burks et al. 2013a; Suppl. material 1: Table S1; Figure 1). The new species, *O. latheticus*, is another similar case. *Oxyscelio latheticus* is extremely similar to *O. convergens* and only differs from the latter by subtle differences, including the shapes of A4, A5 and the metascutellum; however, the *COI* sequences provide strong evidence that these are different species, with a 12.8% maximum interspecific genetic distance between the two species. This example indicates that DNA barcoding is also useful in discovering new species, especially in taxa suspected of harboring cryptic species, such as platygastroid wasps.

Acknowledgements

We are grateful to Drs. Li Ma (Yunnan Agricultural University), Shixiao Luo (South China Botanical Garden) and Chengjin Yan (Wenzhou Vocational College of Science and Technology) for providing fresh specimens for DNA analysis. This study is sup-

ported in part by the National Natural Science Foundation of China (31900346), the Natural Science Foundation of Guangdong Province (2019A1515010588) and the Wencheng County's Second Phase of Innovation and Entrepreneurship Seed Fund in 2019 (2019NKY09).

References

- Burks RA, Masner L, Johnson NF, Austin A (2013a) Systematics of the parasitic wasp genus *Oxytelio* Kieffer (Hymenoptera: Platygasteridae s.l.), Part I: Indo-Malayan and Palearctic fauna. *ZooKeys* 292: 1–263.
- Burks RA, Masner L, Johnson NF, Austin A (2013b) Systematics of the parasitic wasp genus *Oxytelio* Kieffer (Hymenoptera: Platygasteridae s.l.), Part II: the Australian and south-west Pacific fauna. *ZooKeys* 331: 1–266. <https://doi.org/10.3897/zookeys.331.5152>
- Burks RA, Masner L, Johnson NF, Austin AD (2016) Systematics of the parasitic wasp genus *Oxytelio* Kieffer (Hymenoptera, Platygasteridae s.l.), part III: African fauna. *ZooKeys* 565: 29–71. <https://doi.org/10.3897/zookeys.565.7185>
- Chen H, Lahey Z, Talamas EJ, Valerio AA, Popovici OA, Musetti L, Klompen H, Polaszek A, Masner L, Austin AD, Johnson NF (2021) An integrated phylogenetic reassessment of the parasitoid superfamily Platygasteroidea (Hymenoptera: Proctotrupomorpha) results in a revised familial classification. *Systematic Entomology* 46: 1088–1113. <https://doi.org/10.1111/syen.12511>
- Chen H, Talamas EJ, Bon M-C, Moore MR (2020) *Gryon ancina* Kozlov & Lê (Hymenoptera: Scelionidae): host association, expanded distribution, redescription and a new synonymy. *Biodiversity Data Journal* 8: e47687. <https://doi.org/10.3897/BDJ.8.e47687>
- Folmer O, Black M, Hoch W, Lutz R, Vrijenok R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3: 294–299.
- Hebert PDN, Cywinska A, Ball SL, Dewaard JR (2003a) Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences* 270: 313–321. <https://doi.org/10.1098/rspb.2002.2218>
- Hebert PDN, Ratnasingham S, Dewaard JR (2003b) Barcoding animal life: cytochrome c oxidase subunit 1 divergences among closely related species. *Proceedings of the Royal Society of London Series B: Biological Sciences* 270: S96–S99. <https://doi.org/10.1098/rsbl.2003.0025>
- Katoh K, Standley DM (2013) MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular Biology and Evolution* 30: 772–780. <https://doi.org/10.1093/molbev/mst010>
- Kumar S, Stecher G, Tamura K (2016) MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution* 33: 1870–1874. <https://doi.org/10.1093/molbev/msw054>

- Kononova SV, Fursov VN (2007) A review of the genera *Calotelea*, *Calliscelio*, and *Oxyscelio* (Scelioninae, Scelionidae, Proctotrupoidea) from the Palaearctic fauna. Entomological Review 87: 92–105. <https://doi.org/10.1134/S0013873807010101>
- Liao Y-L, Yang B, Xu M-F, Lin W, Wang D-S, Chen K-W, Chen H-Y (2019) First report of *Telenomus remus* parasitizing *Spodoptera frugiperda* and its field parasitism in southern China. In: Talamas E (Ed.) Advances in the Systematics of Platygastridae II. Journal of Hymenoptera Research 73: 95–102. <https://doi.org/10.3897/jhr.73.39136>
- Mo W-h, Chen H-y, Johnson NF, Pang H, Ma L, Liu J-x (2020) Revision of the genus *Oxyscelio* Kieffer (Hymenoptera, Scelionidae) from China. Zootaxa 4816(3): 251–310. <https://doi.org/10.11646/zootaxa.4816.3.1>
- Taekul C, Valerio AA, Austin AD, Klompen H, Johnson NF (2014) Molecular phylogeny of telenomine egg parasitoids (Hymenoptera: Platygastridae s.l.: Telenominae): evolution of host shifts and implications for classification. Systematic Entomology 39: 24–35. <https://doi.org/10.1111/syen.12032>
- Talamas EJ, Bon M-C, Hoelmer KA, Buffington ML (2019) Molecular phylogeny of *Trissolcus* wasps (Hymenoptera, Scelionidae) associated with *Halyomorpha halys* (Hemiptera, Pentatomidae). In: Talamas E (Ed.) Advances in the Systematics of Platygastridae II. Journal of Hymenoptera Research 73: 201–217. <https://doi.org/10.3897/jhr.73.39563>

Supplementary material I

Table S1. Genetic distances between *COI* sequences of *Oxyscelio* from southern China

Authors: Wen-hui Mo, Hua-yan Chen, Hong Pang, Jing-xian Liu

Data type: Doc file

Explanation note: Pairwise distances of 49 *COI* sequences of 22 *Oxyscelio* species.

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