RESEARCH ARTICLE



## A new species of the genus Ambrostoma Motschulsky (Coleoptera, Chrysomelidae, Chrysomelinae) from South Korea, with larval descriptions and biological notes

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#### Abstract

Ambrostoma koreana **sp. n.** is described from South Korea. Detailed descriptions and illustrations of adult and all larval instars are provided and differences to closely related species are discussed. Ovoviviparity is reported for the first time in the genus *Ambrostoma*. Notes on distribution, host plant and biology are also provided.

## Keywords

Chrysomelidae, Ambrostoma, new species, larva, biology, ovoviviparity, Korea

#### Introduction

The genus Ambrostoma was described by Motschulsky (1860) for three species from Russia (Amur region), China and Nepal, respectively, with A. quadriimpressum (Motschulsky, 1845) [= A. superbum (Thunberg, 1787)] as its type species. Since then, nine species have been described in East Asia and the Himalayas (Achard 1922, Chen 1934, 1936, Gressitt and Kimoto 1963, Wang 1992, Kimoto and Osawa 1995, Medvedev 2007). Chen (1936) divided the genus into two subgenera, Ambrostoma and Parambrostoma, based on elytral punctures, setae on elytral epipleura and length ratio of antennomeres, and later Wang and Chen (1981) raised these subgenera to the generic status. These two genera were recently revised to include fifteen species and the monophyly of both genera was well supported by morphological characters of adults (Ge et al. 2012). Ambrostoma is widely distributed in East Asia and comprises eight species. Members of the genus are characterized by a distinct transverse depression near the base of elytra, inner margin of elytral epipleura with setae along an entire length, procoxal cavities open posteriorly, metasternal process immarginate apically and tarsal claws simple. The larva are only known and for only one species of the genus: A. superbum which is a major pest in China, has been described by Medvedev and Voronova (1976). Ambrostoma larva is easily recognized among the subfamily Chrysomelinae by their orange stripes in live specimens, undeveloped tubercles, a dark pattern on the dorsum and black markings on the head and pronotum.

In 2006, the first author collected unusual *Ambrostoma* specimens, adults and larvae, on Namhaedo Island in South Korea. After a detailed examination we concluded that they belong to a new species described below. We compared the larval morphology of *Ambrostoma* to those of *Parambrostoma*. Notes on distribution, host plant, biology and occurrence of ovoviviparity are also provided.

#### Material and methods

The type specimens were deposited in the Department of Biodiversity and Evolutionary Taxonomy, University of Wrocław, Poland (DBET) and H.-W. Cho's private collection, Republic of Korea (HWC). Biological observations were made in April–May 2006, at the type locality and under laboratory conditions. Adults and larvae collected from the type locality were kept in plastic containers (10 cm diameter, 12 cm deep) with leaves of host plant and checked daily. All larval specimens used in the study were preserved in 70% ethanol. For morphological studies of minute structures, some larvae were dissected, cleared in 10% sodium hydroxide solution, rinsed in distilled water, and then mounted on slides with Swan's liquid (20 g distilled water, 15 g gum arabic, 60 g chlorhydrate, 3 g glucose, and 2 g glacial acetic acid). Genitalia were dissected from adult specimens softened in plastic containers with wet tissue paper for 12–24 hours. The aedeagus was softened in 10% sodium hydroxide solution for 6–12 hours and placed in distilled water. The careful insertion of a sharp-pointed thick nose hair and injection of 5% ethanol into the foramen of aedeagus were repeated until the internal sac was fully everted. After washing with absolute ethanol, the genitalia were preserved in a microvial with glycerin and pinned with the specimen. Descriptions and illustrations were prepared using Nikon SMZ800 and Nikon Eclipse E600 microscopes, each equipped with a drawing tube. Habitus images were taken by a Nikon Coolpix 4500 digital camera attached to a Nikon SMZ1500 microscope. The letters L, S and M in parentheses signify long, short, and micro setae, respectively.

#### Results

#### Ambrostoma koreana Cho & Borowiec, sp. n.

http://zoobank.org/4FEAAE6E-87B0-4AFA-B40F-3DAADB68CF75 http://species-id.net/wiki/Ambrostoma\_koreana Figs 1–20

**Type locality.** South Korea: Gyeongnam Province, Namhaedo Island, Mangunsan Mountain, 34°51'52"N, 127°51'47"E.

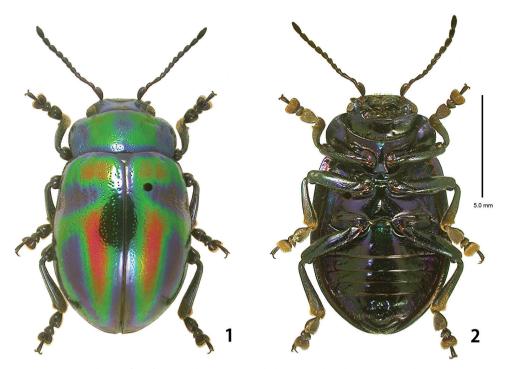
**Type material.** Holotype: male (DBET), KOREA: Gyeongnam Province, Namhaedo Island, Mangunsan Mountain, 34°51'52"N, 127°51'47"E 280 m, 7.IV.2006, H.-W. Cho. Paratypes: 2 males and 6 females (DBET), same data as holotype; 10 males and 10 females (HWC), same data as holotype except for 13.VI.2007, H.-Y. Kwon; 1 male (HWC), KOREA: Gyeongnam Province, Miryang, Gajisan Mountain, 35°35'46"N, 128°59'38"E 500 m, 17.VIII.2001, T.-H. Ahn; 1 female (HWC), same data as preceding paratype except for 25.V.2001, G.-S. Jung.

**Other material.** 42 larvae collected or obtained from adults, same data as holotype except for 23.IV.–15.V.2006.

Adult (Figs 1–7, 19).

**Diagnosis.** The new species is very similar in coloration to *A. leigongshana* Wang, but can be distinguished by the following characters: antennomere 3 much longer than 4 (equal in length in *A. leigongshana*); pronotum with moderately dense punctures in lateral depression (dense and coarse punctures in *A. leigongshana*); aedeagus widest at apical 1/5, thence narrowed with trapezoidal apex (elongate apically with rounded apex in *A. leigongshana*); spermatheca almost straight, curved at apex (strongly curved in *A. leigongshana*).

**Description. Holotype** (Figs 1–2). Body length 11.7 mm, width 5.7 mm, strongly convex dorsally. Head emerald green to ultramarine with 2 pairs of small orange yellow markings on central part. Mouthparts midnight blue. Antennomeres 1–7 ultramarine, 8–11 black. Pronotum emerald green to ultramarine with a pair of large orange yellow markings. Scutellum ultramarine. Elytra ultramarine with a pair of orange yellow markings at base and 2 pairs of orange yellow longitudinal markings on centralposterior part, all markings surrounded by emerald green. Venter mainly ultramarine with greenish luster. Legs midnight blue.



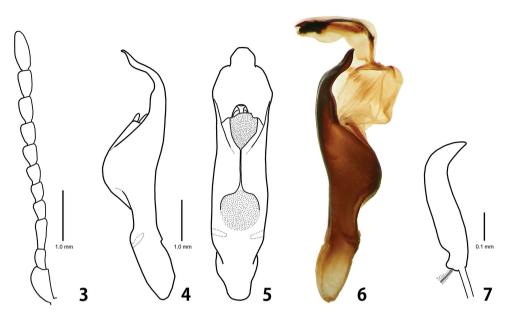
Figures 1-2. Habitus of Ambrostoma koreana sp. n., holotype. I dorsal view 2 ventral view.

*Head.* Width 3.4 mm, interocular distance 2.5 mm. Vertex and frons with sparse and small punctures. Clypeus and labrum with small punctures bearing long setae. Mandibles with moderately dense punctures bearing setae on outer surface. Maxillary palp 4-segmented with apical palpomere distinctly widened and truncate. Antennae (Fig. 3) reaching elytral humeri; antennomere 1 robust, longer than 3; antennomere 2 as long as 4; antennomere 3 distinctly longer than 4; antennomeres 5–10 moderately widened; antennomere 11 longest and 2.6 times as long as wide.

*Pronotum*. Length 2.7 mm, width 5.4 mm. Lateral sides roundly widened anteriorly, widest at anterior 1/4. Anterior margin widely emarginated. Trichobothria on anterior and posterior angles. Disc with dense and small punctures, larger than those of head; hardly confluent large punctures in lateral longitudinal depression; interspaces with spare and minute punctures. Scutellum subtriangular, slightly wider than long, impunctate.

*Elytra.* Length 8.8 mm, width 6.8 mm. Sides moderately widened posteriorly and widest at posterior 2/5, thence rounded at apex. Disc with double irregular rows of punctures; transverse depression with large punctures, subequal to those of side of pronotum; interspaces with spare and minute punctures. Epipleuron flat, inner margin with micro setae along an entire length. Hind wing well developed.

*Venter.* Hypomera impunctate; prosternum with sparse and small punctures; prosternal process strongly enlarged and slightly emarginated apically. Abdominal sternites wrinkled laterally with sparse and small punctures; last abdominal sternite deeply emar-



Figures 3–7. Adult of *Ambrostoma koreana* sp. n. 3 antenna 4 aedeagus lateral view 5 aedeagus dorsal view 6 aedeagus with everted internal sac laternal view 7 spermatheca.

ginated on both sides. Legs moderately robust; tibiae simple without preapical tooth; fore and middle legs with tarsomere 1 slightly narrower than 3; tarsal claws simple.

*Aedeagus*. Strongly convex at middle, curved and sinuated at apex in lateral view (Fig. 4); subparallel-sided, widest at apical 1/5, thence narrowed with trapezoidal apex, with 2 weakly sclerotized plates in dorsal view (Fig. 5); internal sac curved, shorter than median lobe with thick and long flagellum (Fig. 6).

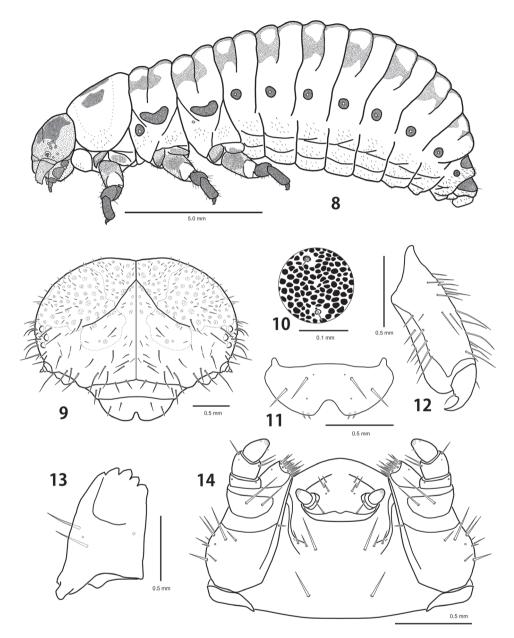
**Paratypes.** Body length 10.9–12.7 mm, width 5.2–6.2 mm. Coloration similar to holotype. Female: larger than male; tarsomere 1 of fore and middle legs distinctly narrower than 3; last abdominal sternite rounded; spermatheca (Fig. 7) almost straight, curved at apex.

#### First instar larva (Figs 15, 17).

**Description.** Body length 2.7-4.2 mm, width 1.2-1.7 mm, head width 0.90-0.95 mm (n = 5). Body convex dorsally on abdomen. Pale yellow with head dark brown, tubercles and legs brown (Fig. 17). Tubercles weakly developed (Fig. 15). Sclerotized platelets on dorsum dense and strong, on venter almost absent. Setae longer than in other instars, bases of setae sclerotized.

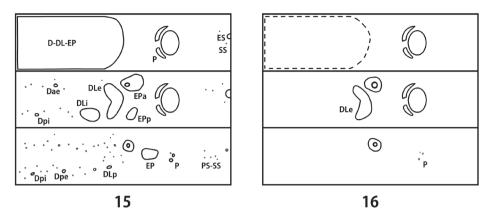
*Head.* Vertex and temporal side with 30–32 pairs of long setae and 15–19 pairs of short setae. Frons with 16–19 pairs of long setae. Clypeus and labrum, each with 2 pairs of long setae. Mouthparts similar in shape and chaetotaxy to those of the last instar larva, except for stipes with 3 setae and postmentum with 3 pairs of setae.

*Thorax.* Prothorax with D-DL-EP (53–57L 4–6S); P (1L); ES (1L) weakly sclerotized; SS represented by a short seta; sternal region with 1–2 additional setae. Meso-



Figures 8–14. Last instar larva of *Ambrostoma koreana* sp. n. 8 habitus 9 head 10 sclerotized platelets on dorsal mesothorax 11 labrum 12 tibia and tarsungulus 13 mandible 14 lower mouthparts.

and metathorax with Dae (1L); Dpi (1L); dorsal region with 11–13 additional short setae; DLi (1–3L) well developed with egg burster; DLe (4–5L 8–11S); EPa (4–5L 5S) fused with spiracle; EPp (2L 3–4S); P (1L); ES (1L) weakly sclerotized; SS represented by a short seta; sternal region with 2–3 additional setae.



**Figures 15–16.** Schematic presentation of tubercular patterns (top: prothorax, middle: mesothorax, bottom: 2nd abdominal segment). **15** first instar larva **16** last instar larva.

*Abdomen.* Dorsal and dorsolateral regions with 31–35 short setae; Dpi (1L); Dpe (1L); DLp (1L); EP (9–10L 8–10S); P (3S) divided; PS-SS represented by 4–5 short setae; ES (1S); segment 8 with D-DL (3–4L 11–12S) fused; segment 9 with D-DL-EP (3L 11–12S) fused; segment 10 with pygopod developed; egg burster on segment 1 much smaller than thoracic ones.

#### Second instar larva.

**Description.** Similar to the last instar larva except for following characters: body length 4.2-5.7 mm, width 1.8-2.4 mm, head width 1.25-1.35 mm (n = 4); black markings on head and dark patterns on dorsum much larger than those of the last instar larva; pronotum with black markings partly connected.

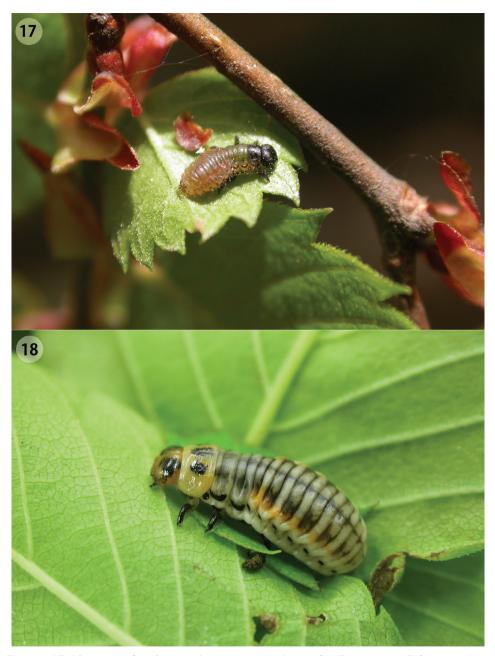
## Third instar larva.

**Description.** Similar to the last instar larva except for following characters: body length 7.2–8.1 mm, width 2.8–3.8 mm, head width 1.90–2.05 mm (n = 3); dark patterns on dorsum much larger than those of the last instar larva.

Fourth (last) instar larva (Figs 8–14, 16, 18).

**Diagnosis.** The last instar larva is easily distinguished from larva of *A. superbum* (Thunberg) in the following characters: pronotum with 3 black markings (2 black markings in *A. superbum*); dorsolateral posterior region of each abdominal segment without dark patterns (with dark patterns in *A. superbum*); width of peritreme less than half width of abdominal segment (equal to half width in *A. superbum*).

**Description.** Body length 11.6–12.8 mm, width 4.7–5.3 mm, head width 2.60–2.65 mm (n = 5). Body strongly convex dorsally on abdomen (Fig. 8). Yellowish white in alcohol specimens, creamy white with orange stripes in live specimens (Fig. 18). Head yellowish brown with a pair of large black markings. Pronotum with 3 black markings. Tubercles, spiracles and legs dark brown. Dorsum with dark patterns consisting of dense and strong sclerotized platelets, sparsely covered with micro setae (Fig. 10). Tubercles undeveloped (Fig. 16). Venter covered with short and moderately long setae, bases of setae not sclerotized.



Figures 17–18. Larvae of *Ambrostoma koreana* sp. n. on leaves of *Zelkova serrata*. 17 first instar larva 18 last instar larva.

*Head.* Hypognathous, rounded, well sclerotized, covered with dark spots (Fig. 9). Vertex and temporal side with 10–11 pairs of long setae and 77–82 pairs of short setae. Epicranial suture Y-shaped; coronal suture distinct along an entire length;



Figure 19. Overwintered adults of Ambrostoma koreana sp. n.

frontal suture indistinct for lateral 2/5. Frons slightly depressed medially with 14–15 pairs of long setae and 17–22 pairs of short setae. Endocarina distinct; epistomal suture developed. Six stemmata on each side. Antenna 3-segmented; antennomere 1 without seta; antennomere 2 with a sensory papilla and 4 setae; antennomere 3 with 5 setae. Clypeus trapezoid with 2 pairs of setae. Labrum (Fig. 11) deeply emarginate anteriorly with 2 pairs of setae; epipharynx with 2 pairs of setae. Mandibles (Fig. 13) symmetrical, 5-toothed with 2 setae. Maxillary palp (Fig. 14) 3-segmented; palpomere 1 without seta; palpomere 2 with 3 setae; palpomere 3 conical with a seta; palpiger with 2 setae; mala with 13–14 setae; stipes with 10–13 setae; cardo without seta. Labial palp 2-segmented; prementum with 4–5 pairs of setae.

*Thorax.* Prothorax with D-DL-EP scattered with micro setae, not sclerotized; P (6–7S); ES and SS, each represented by a short seta; sternal region with 14–16 additional setae. Meso- and metathorax with DLe (27–30M); P (8–10S); EPa and EPp, each represented by 23–27 short setae; ES and SS, each represented by a long seta; sternal region with 20–35 additional setae; mesothoracic spiracle annuliform with large peritreme; metathoracic spiracle vestigial. Legs rather stout; tibia with 28–30 setae; tarsungulus strongly curved; basal tooth weakly developed with a seta (Fig. 12).

*Abdomen.* Tubercles absent on segments 1–7 except for P represented by 3–4 short setae arising from sclerotized bases; epipleural to sternal regions with a lot of short setae. Segment 8 with D-DL (10–12S); segment 9 with D-DL-EP (12–14L 1–2M); segment 10 with pygopod well developed; spiracle with large peritreme similar to mesothoracic one, but smaller; eversible glands absent.

Etymology. This endemic species is named after the type locality, Korea.

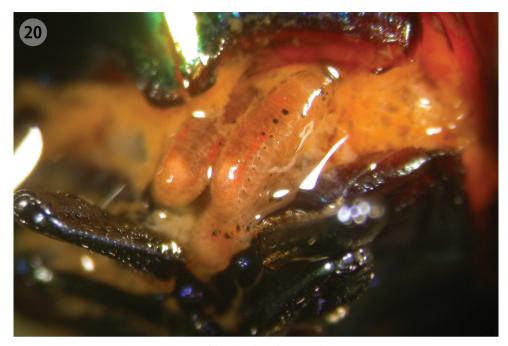


Figure 20. Larvae inside the abdomen of female of Ambrostoma koreana sp. n.

**Distribution.** Specimens were collected in the southeastern part of the Korean Peninsula and on adjacent Namhaedo Island.

**Biological notes.** Overwintered adults were observed under bark or in fallen leaves near the base of trees in early April (Fig. 19). Adults and larvae fed on leaves of *Zelkova serrata* Makino (Ulmaceae) which is quite a common tree in Korea. *Ulmus pumila* Linnaeus (also Ulmaceae) was the only known host plant for three other *Ambrostoma* species (Yu et al. 1996). On 7 May 2006, a female laid 4 eggs that contained fully developed embryos in the laboratory. Four females were dissected shortly afterward, and 17–79 enclosed larvae were found in abdomen (Fig. 20). About 50 ovoviviparious species are actually known in 7 genera of Chrysomelinae (Bontems 1988, Bontems and Lee 2008). Ovoviviparity is reported for the first time in the genus *Ambrostoma*.

#### Key to the adults of Ambrostoma species (modified from Ge et al. 2012)

1	Punctures of transverse impression of elytra subequal to those of the lateral
	sides of pronotum
_	Punctures of transverse impression of elytra much finer than those of sides of
	pronotum
2	Elytra with 3 broad transverse bands of purplish copper-red

_	Elytra with 1 transverse and 4 longitudinal bands of orange yellow
3	Elytra with large well-defined post-median violaceous patch surrounded by
	green4
-	Elytra without well-defined post-median violaceous patch surrounded by
	green
4	Arrangement of elytral punctures almost entirely irregular; last abdominal
	sternite of male deeply emarginate on both sidesA. fulgurans (Achard)
_	Elytral punctures arranged in simple rows at base before transverse impres-
	sion, slightly irregular beyond impression; last abdominal sternite moderately
	sinuate on both sides
5	Pronotum slightly dilated in front of middle region; interspaces between ely-
	tral striae finely and densely punctuate
_	Pronotum strongly dilated in front of middle region; interspaces between
	elytral striae finely and very sparsely punctuate
6	Elytra with single striae A. chinkinyui Kimoto & Osawa
_	Elytra with double striae
7	Pronotum with dense and coarse punctures in lateral depression
_	Pronotum with sparse punctures in lateral depression
	A. omeishanum Gressitt & Kimoto
8	Elytra with longitudinal purple markings on media posterior surrounded by
	green; last abdominal segment in male shallowly sinuate
	A. leigongshanum Wang
	8 8 8
_	Elytra with 4 purple longitudinal stripes on media posterior part; last ab-
	dominal segment in male deeply sinuate

## Discussion

The Ambrostoma species has not been recorded in Korea since Chen (1936) mentioned A. superbum without any locality data and comments. Ambrostoma superbum is widely distributed in Southeastern Siberia, Eastern Mongolia, Northeastern China and probably North Korea (Kippenberg 2010, under name A. quadriimpressum quadriimpressum), whereas A. koreana is restricted to Namhaedo Island and Miryang, Gyeongnam Province, South Korea. The first instar larva of Ambrostoma was described for the first time. The second and third instar larva are very similar in shape and coloration to the last instar larva, but the first instar larva distinctly differs in having more developed tubercles and long setae. The tubercular pattern of the first instar larva was similar to some larvae of the genus Chrysolina Motschulsky. Based on body shape, tubercular pattern and biology, Ambrostoma larva belongs to the generic group Chrysolina (Takizawa 1989). The monophyly of both genera, Ambrostoma and Parambrostoma carried out by Ge et al. (2012), was supported by larval characters.

The mature larvae of these two genera are similar in having undeveloped dorsal tubercles, sclerotized platelets and sparse micro setae on dorsum. However, the considerable differences can be summarized as follows: dark patterns present on dorsum in *Ambrostoma* and absent in *Parambrostoma*; head and pronotum are pale coloured with black markings in *Ambrostoma* and entirely black in *Parambrostoma*; prothoracic and ventral tubercles are not scleotized in *Ambrostoma*, whereas they are well scleotized in *Parambrostoma*; basal tooth of tarsungulus in *Ambrostoma* is weakly developed while in *Parambrostoma* it is strongly developed.

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## References

- Achard J (1922) Descriptions de nouveaux Chrysomelini (Col. Chrysomelidae). Fragments Entomologiques 1–2: 13–28.
- Bontems C (1988) Localization of spermatozoa inside viviparous and oviparous females of Chrysomelinae. In: Jolivet P, Petitpierre E, Hsiao T (Eds) Biology of Chrysomelidae. Kluwer Academic Publishers, Dordrecht, 299–316. doi: 10.1007/978-94-009-3105-3\_18
- Bontems C, Lee CF (2008) A new case of viviparity among Chrysomelinae. In: Jolivet P, Santiago-Baly J, Schmitt M (Eds) Research on Chrysomelidae, Vol 1. Brill, Leiden-Boston, 260–264.
- Chen SH (1934) On some species of Chrysomelidae (Col.) in the British Museum. Stylops 3: 66–78.
- Chen SH (1936) The chrysomelid genus Ambrostoma Motsch. Sinensia 7 (6): 713-729.
- Ge SQ, Daccordi M, Beutel RG, Ren J, Cui JZ, Li WZ, Yang XK (2012) Revision of the Eastern Asian genera Ambrostoma Motschulsky and Parambrostoma Chen (Coleoptera: Chrysomelidae: Chrysomelinae). Systematic Entomology 37: 332–345. doi: 10.1111/j.1365-3113.2012.00618.x
- Gressitt JL, Kimoto S (1963) The Chrysomelidae (Coleopt.) of China and Korea, Part 2. Pacific Insect Monograph 1B: 301–1026.
- Kimoto S (1962) A phylogenic consideration of Chrysomelinae based on immature stages of Japanese species (Coleoptera). Journal of the Faculty of Agriculture, Kyushu University 12 (2): 67–88.
- Kimoto S, Osawa S (1995) Description of a new species of the genus *Ambrostoma* Motschulsky from Taiwan, China (Coleoptera, Chrysomelidae, Chrysomelinae). Entomological Review of Japan 50: 15–16.

- Kippenberg H (2010) Subfamily Chrysomelinae. In: Löbl I, Smetana A (Eds) Catalogue of Palaearctic Coleoptera, Vol. 6 Chrysomeloidea. Apollo Books, Stenstrup, 390–443.
- Medvedev LN (2007) New taxa of Oriental Chrysomelidae (Coleoptera). Euroasian Entomological Journal 6: 433–438.
- Medvedev LN, Voronova NV (1976) A description of the larva of *Ambrostoma* (Chrysomelidae) and the distribution of the genus *Ambrostoma* in Mongolia. In: Kerzhner IM (Ed) Insects of Mongolia, Issue 4. Nauka, Leningrad, 237–240.
- Motschulsky V (1860) Coléoptères de la Sibérie orientale et en particulier des rives de l'Amour. In: Schrenck L von: Reisen und Forschungen im Amur-Lande in den Jahren 1854–1856. Vol. 2, part 2. L. v. Schrenk, St. Petersburg, 77–258, plates VI–XI.
- Takizawa H (1989) Notes on larvae of the subfamily Chrysomelinae (Coleoptera, Chrsyomelidae), part 1. Kanagawa-chuho 90: 243–256.
- Wang SY (1992) Two new species of leaf beetles from Wuling mountain of China (Coleoptera: Chrysomelidae). Sinozoologia 9: 175–178.
- Wang SY, Chen SC (1981) Coleoptera: Chrysomelidae: Chrysomelinae. In: Huang FS (Ed) Insects of Xizang, Vol. 1. Science Press, Beijing, 509–516.
- Yu PY, Wang SY, Yang XK (1996) Economic Insect Fauna of China, Fasc. 54, Coleoptera: Chrysomeloidea (II). Science Press, Beijing, 324 pp.

RESEARCH ARTICLE



## Description of two new species of the leafhopper subgenus Pediopsoides (Pediopsoides) (Hemiptera, Cicadellidae, Macropsinae) from Guangxi Province, Southern China

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## Abstract

Two new species of the Macropsinae leafhopper subgenus *Pediopsoides* (*Pediopsoides*) Matsumura, 1912, *P. (P.) damingshanensis* Li, Dai & Li, **sp. n.** and *P. (P.) tishetshkini* Li, Dai & Li **sp. n.**, are described and illustrated from Guangxi Province of southern China. A key to males is provided to distinguish the species of the subgenus along with a map showing the distribution of the new species.

## Keywords

Auchenorrhyncha, taxonomy, morphology, description

## Introduction

The leafhopper genus *Pediopsoides* (Insecta: Auchenorrhyncha: Membracoidea: Cicadellidae: Macropsinae) was established by Matsumura (1912) for a single species, *Pediopsoides formosanus* Matsumura, 1912, from Taiwan island of China. In Hamilton's (1980) Macropsini revision the genus *Pediopsoides* included 5 subgenera, including the nominate subgenus *Pediopsoides* (*Pediopsoides*) for *P*. (*P*.) formosanus Matsumura, 1912 from Taiwan island of China and *P*. (*P*.) satsumensis (Matsumura, 1912) from Kiushu island of Japan. More recently, the following other species have been added: *P*. (*P*.) femorata (Hamilton, 1980) from Taiwan island of China, *P*. (*P*.) kodaiana Viraktamath, 1996 from Tamil Nadu state of India, *P*. (*P*.) jin-gdongensis Zhang, 2010 from Yunnan Province of China, *P*. (*P*.) bispinata Li, Dai & Li, 2012 and *P*. (*P*.) nigrolabium Li, Dai & Li, 2012 from Guangxi Province of China. Until now there are seven species of the nominate subgenus including five from China.

In the present paper, 2 new species, *Pediopsoides* (*Pediopsoides*) damingshanensis Li, Dai & Li sp. n. and P. (P.) tishetshkini Li, Dai & Li sp. n., from Guangxi province (included in the oriental region) of southern China are described and illustrated, a key to species of the nominate subgenus are provided, along with a map showing the distribution of the new species.

## Material and methods

Terminology used in describing the structures follows Hamilton (1980).

External morphology and dissected male genitalia were observed under an Olympus SZ2-ILST stereoscopic microscope and YS 100 microscope. Habitus images of adults were obtained by using a KEYENCE VHX-1000 system and were processed using the software Adobe Photoshop CS6. Hand drawings of the male genitalia were processed with Adobe Illustrator CS6. The body length is measured from the apex of the head to the apex of the forewings and are given in millimeter (mm.).

The type specimens of the new species are all deposited in the Institute of Entomology, Guizhou University, Guiyang, China (GUGC).

#### Taxonomy

## Subgenus Pediopsoides (Pediopsoides) Matsumura

*Pediopsoides* Matsumura, 1912: 305. *Pediopsoides* (*Pediopsoides*) Hamilton, 1980: 896.

#### Type species. Pediopsoides formosanus Matsumura, 1912, by original designation.

**Remarks.** The nominate subgenus was characterized by Hamilton (1980) and Li et al. (2012). It can be distinguished by the following combination of features: pronotal striations oblique, forewings with two anteapical cells, dorsal connective freely attached (fused in subgenus *P*. (*Celopsis*)) to tenth tergite, and male pygofer without multifid or additional spines [male pygofer spine bifid or with rounded lobe on inner margin

basally in subgenus *P*. (*Sispocnis*), a small secondary pygofer spine posteriorly in subgenus *P*. (*Nanopsis*), and male pygofer spine are multifid in subgenus *P*. (*Kiamoncopsis*)].

Distribution. Oriental region: China, Japan and India.

*Pediopsoides (Pediopsoides) damingshanensis* Li, Dai & Li, sp. n. http://zoobank.org/8874F63A-824B-43CC-9509-F1D6180A8B5E http://species-id.net/wiki/Pediopsoides\_damingshanensis Figures 1–3, 7–13, 22

Type locality. CHINA: Guangxi Province, Damingshan.

**Measurements.** Body length (including tegmina),  $\mathcal{E}$ , 4.0 mm.

**Description.** *Body coloration.* General color (Figs 1–3) yellowish brown. Face (Fig. 3) yellowish with slight greenish tinge, eyes pale brown, slight brown oblique band adjacent inner sides of ocelli, lower part of face pale brown. Pronotum (Fig. 1) dark brown centrally, lateral and front margins gradually more yellowish. Scutellum (Fig. 1) dark brown except lateral margins yellowish. Forewings (Fig. 2) evenly dark brown except yellowish basal part. Legs pale yellowish.

*External morphology.* Head, face, pronotum, and scutellum faintly striated. Head (Fig. 1), in dorsal view, clearly arcuate forward, slightly narrower than pronotum, crown distinctly shorter medially than next to eyes. Face (Fig. 3), in lateral view, moderately flat; in facial view, as wide as long across eyes; dorsal part of frontoclypeus with slight medial longitudinal carina; ocelli about 9 times their diameter to adjacent eye; lacking clear sutures between lora and frontoclypeus. Pronotum (Fig. 1) 2.2 times as long as wide, frontally and laterally with oblique striations; anterior margin strongly arched, posterior margin excavate medially. Scutellum (Fig. 1) 1.3 times longer than pronotum. Forewings (Fig. 2) hyaline, with 2 anteapical cells, venation protruding. Hind femoral macrosetae 2+1; hind tibia with 7 macrosetae on AD row, 5 on AV row, 11–12 on PD row, dense and slender on PV row.

*Male genitalia*. Pygofer (Fig. 7), with dorsal margin incised, caudal margin truncate, slightly sinuated in lateral view, ventral margin serrate distally with few fine setae. Subgenital plate (Fig. 7), in lateral view, slightly shorter than pygofer, slender, rodlike, with many scattered setae. Style (Fig. 8), slender, apophysis margined with short fine setae, slightly angled after lateral lobe, slightly inflated subapically, apex subacute. Connective (Figs 9–10) distinctly longer than greatest width basally, with anterior medial process long, arms bent dorsally. Aedeagus (Figs 11–12), with short preatrium; dorsal apodeme moderately long; shaft sinuate, in lateral view apex truncate, in ventral view broad basally, widened subapically; gonopore long, apical on ventral margin. Dorsal connective (Fig. 13) "S" shaped with acute apex.

Female. Unknown.

**Type material.** Holotype, ♂, CHINA: Guangxi Province, Damingshan National Natural Reserve, 14. V. 2012, collected by Li Hu.

Distribution. Guangxi Prov. (Damingshan), China (Fig. 22).



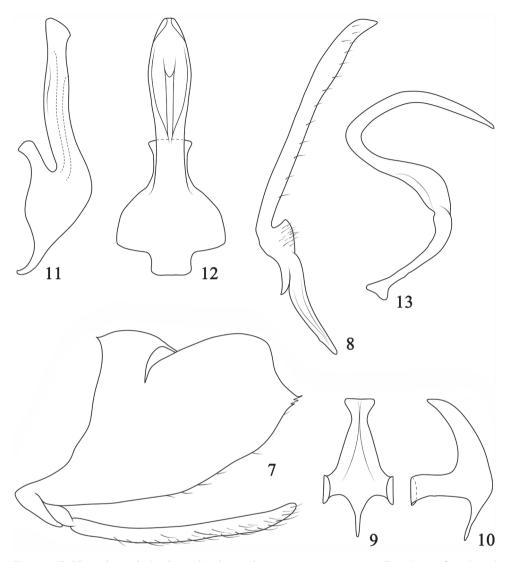
Figures 1–6. 1–3 *Pediopsoides (Pediopsoides) damingshanensis* Li, Dai & Li sp. n. 4–6 *P. (P.) tishetshkini* Li, Dai & Li sp. n. 1, 4 Dorsal habitus, male 2, 5 Lateral habitus, male 3, 6 Face.

**Diagnosis.** This new species differs from other members of the subgenus *Pediopsoides* (*Pediopsoides*) by the shape of the male genitalia.

Etymology. The new species name refers to the type locality, Damingshan.

*Pediopsoides (Pediopsoides) tishetshkini* Li, Dai & Li, sp. n. http://zoobank.org/9A8FCAC5-D112-43BC-A401-367DCDCE83E9 http://species-id.net/wiki/Pediopsoides\_tishetshkini Figures 4–6, 14–22

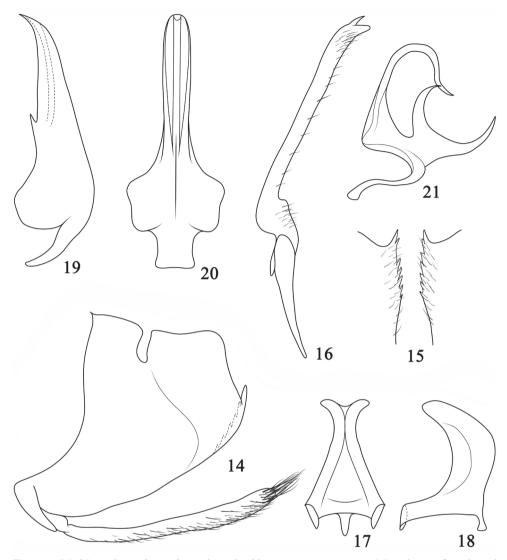
**Type locality.** CHINA: Guangxi Province, Tianlin. **Measurements.** Body length (including tegmina), ♂, 4.5 mm.



Figures 7–13. *Pediopsoides (Pediopsoides) damingshanensis* Li, Dai & Li sp. n. 7 Male pygofer side and subgenital plate, lateral view 8 Style, dorsal view 9 Connective, dorsal view 10 Connective, lateral view 11 Aedeagus, lateral view 12 Aedeagus, ventral view 13 Dorsal connective, lateral view.

**Description.** Body coloration. Color (Figs 4–6) similar to *P*. (*P*.) damingshanensis but darker; with large black spot adjacent antennal pit and basal parts of fore femur and coxa, middle and hind coxa marked with dark brown.

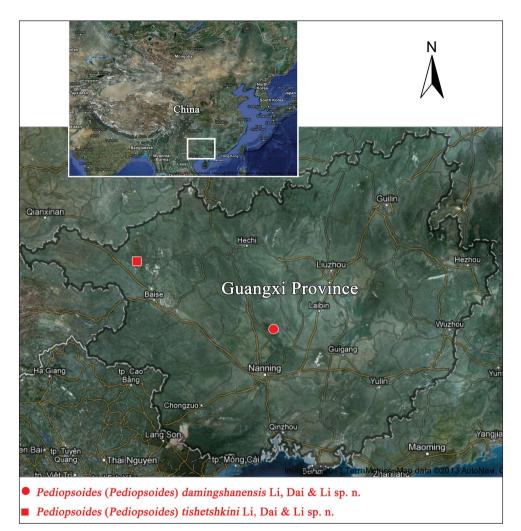
*External morphology.* As in *P.* (*P.*) *damingshanensis* but head more narrower than pronotum and crown more arcuate forward; face (Fig. 6), in lateral view, slightly inflated; ocelli with their spacing about 8 times than that of ocellus to adjacent eye; scutellum (Fig. 4) about 1.4 times longer than pronotum. Hind femoral macrosetae



Figures 14–21. *Pediopsoides (Pediopsoides) tishetshkini* Li, Dai & Li sp. n. 14 Male pygofer side and subgenital plate, lateral view 15 Ventral margins of male pygofer 16 Style, dorsal view 17 Connective, dorsal view 18 Connective, lateral view 19 Aedeagus, lateral view 20 Aedeagus, ventral view 21 Dorsal connective, lateral view.

2+1; hind tibia with 7 macrosetae on AD row, 6 on AV row, 11 on PD row, dense and slender on PV row.

*Male genitalia*. Similar to *P*. (*P*.) *damingshanensis* but pygofer (Fig. 14) shorter, style (Fig. 16) with lateral lobe shorter and apex bearing a spine-like process and connective (Figs 17–18) more robust in lateral view. Aedeagus (Figs 19–20) with basal apodeme reduced; preatrium moderately long; shaft in lateral view relatively straight,



**Figure 22.** Map showing the distribution of *Pediopsoides* (*Pediopsoides*) *damingshanensis* Li, Dai & Li sp. n. and *P*. (*P*.) *tishetshkini* Li, Dai & Li sp. n.

evenly tapered to sharply pointed and upturned apex; in ventral view shaft similar in width throughout length with rounded apex; gonopore apical on ventral margin. Dorsal connective (Fig. 21) strongly developed "S" shaped, medially produced into bifurcate process, two dorsal branches widely spaced and tapered to acute apex.

Female. Unknown.

**Type material.** Holotype, ♂, CHINA: Guangxi Province, Baise City, Tianlin County, Langping Village, 23. IV. 2012, collected by Zheng Weibin.

Distribution. Guangxi Prov. (Tianlin), China (Fig. 22).

**Remarks.** The new species is similar to *P*. (*P*.) *damingshanensis* sp. n. but can be distinguished by its darker colour and differences in the male genitalia (see description).

**Etymology.** This species is named in honour of Dr. Dmitri Yu. Tishechkin for his excellent contribution to *Macropsis* systematics of the Palaearctic region, and invaluable help to the first author.

#### Key to species (males only) of the subgenus Pediopsoides (Pediopsoides)

Male *P*. (*P*.) *formosanus* (Matsumura) is not known, hence not included in the key. The present key is modified from Li et al. 2012.

1	Aedeagal shaft with processes	
_	Aedeagal shaft without processes (Figs 11–12, 19–20)	
2	Aedeagal shaft with apical processes laterally directed and on either side of	
	gonopore	
_	Aedeagal shaft with subapical processes directed dorsally or ventrally3	
3	Aedeagal shaft processes directed dorsally P. (P.) kodaiana Viraktamath	
_	Aedeagal shaft processes directed ventrally P. (P.) femorata (Hamilton)	
4	Aedeagal shaft inflated subapically in lateral view	
_	Aedeagal shaft either of uniform width (Fig. 11) or narrowed subapically	
	(Fig. 19)	
5	Clypellus black	
_	Clypellus not black	
6	Aedeagal shaft of uniform width in lateral view (Fig. 11); dorsal conne	
	S-shaped (Fig. 13) P. (P.) dainghanensis Li, Dai & Li sp. n.	
_	Aedeagal shaft tapered variously distally (Fig. 19)7	
7	Style with subapical tooth-like process (Fig. 16)	
_	Style without a subapical tooth-like process P. (P.) bispinata Li, Dai & Li	

## Acknowledgements

The authors thank Zheng Wei-Bin (Institute of Entomology, Guizhou University, Guiyang, Guizhou, China) for providing material studied. This project was supported by the National Natural Science Foundation of China (31000952).

## References

Hamilton KGA (1980) Contributions to the study of the world Macropsini (Rhynchota: Homoptera: Cicadellidae). The Canadian Entomologist 112: 875–932. doi: 10.4039/ Ent112875-9

- Huang KW, Viraktamath CA (1993) The Macropsine Leafhoppers (Homoptera: Cicadellidae) of Taiwan. Chinese Journal of Entomology 13: 361–373.
- Li H, Dai R-H, Li Z-Z (2012) Leafhopper Subgenus *Pediopsoides* (*Pediopsoides*) (Hemiptera, Cicadellidae, Macropsinae) with Descriptions of Two New Species from Southern China. Florida Entomologist 95 (3): 535–542. doi: 10.1653/024.095.0301
- Matsumura S (1912) Die Acocephalinen und Bythoscopinen Japans. The journal of the Agriculture, Tohoku Imperial University, Sapporo, Japan 4 (7): 279–325.
- Rakitov RA (1998) On differentiation of cicadellid leg chaetotaxy (Homoptera, Auchenorrhyncha, Membracoidea). Russian Entomological Journal 6: 7–27.
- Viraktamath CA (1996) New Oriental Macropsinae with a key to species of the Indian subcontinent (Insecta: Auchenorrhyncha: Cicadellidae). Entomologische Abhandlungen, Stätliches Museum für Tierkunde, Dresden 57 (7): 183–200.
- Zhang B (2010) Two new species of the macropsine leafhopper genus *Pediopsoides* Matsumura from southwest China (Hemiptera: Cicadomorpha: Cicadellidae). Zootaxa 2620: 56–62.

RESEARCH ARTICLE



# A new genus and species of Coenosiini from Costa Rica (Diptera, Muscidae, Coenosiinae)

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http://zoobank.org/249D5395-F5FE-45F6-9DF9-B3535FEBCA3E		

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## Abstract

*Palpilongus* **gen. n**. is herein described for one species – *P. bifurcus* **sp. n**., from Costa Rica, based on male and females. The striking morphological characters of the species – palpus very long, about as long as prementum; upper calypter truncate and very short and setae of male sternite 5 bifurcated, confirm that this new species is also a new genus in the tribe Coenosiini. Male and female terminalia were dissected and illustrated.

## Keywords

Morphology, Neotropical region, Palpilongus gen. n., Palpilongus bifurcus sp. n., taxonomy

## Introduction

The Coenosiinae, a subfamily of the Muscidae, comprises the tribes Limnophorini and Coenosiini, both with genera in the Neotropical region (Couri and de Carvalho 2002). Members of the Coenosiini tribe share: (i) eyes usually dichoptic in both sexes, (ii) lower proepimeral seta downward directed, (iii) katepisternal setae 1:1:1, forming an equilat-

eral triangle, (iv) hind tibia with at least one anterodorsal seta and (v) male hypandrium tubular and elongated (Couri and Carvalho 2002). The tribe is monophyletic, and the arrangment of the katepisternal setae was the single synapomorphy that supported the monophyly of the Coenosiini in Couri and Pont (2000) analysis. A downcurved lower proepimeral setae was first pointed out by Malloch (1934) as another synapomorphy for Coenosiini, but this character also appears in *Agenamyia* Albququerque temporarely transferred by Couri and Pont (2000) to the Limnophorini tribe, of the Coenosiinae subfamily.

Most of the species are known as predators of other insects and some play an important role as potential biocontrol agents, as, for instance, *Coenosia attenuata* Stein (Couri and Salas 2010).

Coenosiini is the largest tribe of Coenosiinae, with 29 genera in the world, 15 in the Neotropical region. Four are currently known from Costa Rica: *Bithoracochaeta* Stein, 1911, *Cordiluroides* Albuquerque, 1954, *Neodexiopsis* Malloch, 1920 and *Schoenomyza* Haliday, 1833 (de Carvalho et al. 2005; Couri et al. 2006).

The aim of the present contribution is to describe a new Coenosiinae species from Costa Rica, and to ascribe it to a new genus based on unique combination of characters.

#### Materials and methods

This study was based on one male and eight females specimens from Costa Rica in the collection of the Instituto Nacional de Biodiversidad (INBIO, Costa Rica). Two female paratypes (one each) will be deposited at the Museu Nacional, Universidade Federal do Rio de Janeiro (MNRJ) and at the Department of Zoology of the Universidade Federal do Paraná (DZUP). Holotype and the remaining paratypes remain at INBIO. Terminology follows McAlpine (1981) except for postpedicel instead of antennal flagellomere, as we followed Stuckenberg (1999).

The terminalia were macerated in a 10% potassium hydroxide solution at room temperature for 24 hours. They were then dissected in glycerol and stored in a microtube with the specimen. Color photos were taken with Syncroscopy, JVC Auto-Montage with a Leica MZ 16 optical microscope.

#### Taxonomy

## Genus Palpilongus gen. n.

http://zoobank.org/40B2A451-4FEA-408A-8509-0FBE48BA61AF http://species-id.net/wiki/Palpilongus

#### Type-species. Palpilongus bifurcus sp. n., by present designation

**Diagnosis.** (Fig. 1) Male dichoptic; eye bare, separated by about 1/3 of head width in both sexes; palpus flattened, very long with length equal to that of the prementum (Fig. 2); proepimeral seta oriented downwards; notum and pleurae with very few setu-



Figure 1. Palpilongus bifurcus gen. n. et sp. n. (holotype), habitus in lateral view.

lae; presutural acrostichal setae developed; dorsocentral setae 1 + 3; katepisternals 1 + 1 + 1 forming an equilateral triangle; upper calypter truncate and very short; wing veins bare; male hind tibia with many rows of fine and long anterodorsal, dorsal and posterodorsal setae; sternite 1 bare; setae on sternite 5 bifurcated; hypandrium tubular in male; ovipositor long in female.

**Etymology.** Derived from the Latin words *palpus* and *longus*, the genus name refers to the long palpus.

**Discussion.** In the current classification of Muscidae, the new genus falls in the tribe Coenosiini of the Coenosiinae. In both keys to muscid genera identification of de Carvalho and Couri (2002) for the Neotropical region and Savage and Vockeroth (2010) for Central America, the new genus approaches *Neodexiopsis* and can be separated by the following couplet:

 Palpus short, much shorter than prementum length; upper calypter glossiform and not very short; setae on sternite 5 not bifurcated ... *Neodexiopsis* Malloch
 Palpus very long about as long as prementum; upper calypter truncate and very short; setae on sternite 5 bifurcated ......*Palpilongus* gen. n.

The new genus was added to the cladistic analysis of the world Coenosiini (Couri and Pont 2000). The analysis positioned *Palpilongus* in the same clade of *Cordiluroides* 

+ *Neodexiopsis* + *Haroldopsis* Albuquerque (synonymyzed with *Neodexiopsis* in the referred cladistic analysis), based on one synapomorphy - presence of three preapical setae on mid femur. The new genus was supported by the following synapomorphies: hairs on arista at most equal to basal width of arista; fronto-orbital plate with no setulae; colour of thorax and abdomen shinning undusted; notum almost bare, with very few ground setulae; lateral seta on scutellum present and hind tibia with a preapical posterodorsal seta at apical fourth.

*Neodexiopsis* and *Cordiluroides* are differently represented in the Neotropical region. *Neodexiopsis* is a speciose muscid genus, with about hundred described species found throughout the region, while *Cordiluroides* have a more restricted geographic distribution (Mexico, Costa Rica and Brazil) and is known by 6 species.

Most of the *Neodexiopsis* species have light brown to dark brown bodies, grey pollinose, and yellow to brown legs. Adults are small to medium sized predatory flies that inhabit forests or open habitats. Similarly to other genera of the Coenosiini tribe, the characteristic chaetotaxy of the hind tibia can distinguish one from each other. *Neodexiopsis* species can be recognized by the presence of one anterodorsal, one posterodorsal and one anteroventral setae in hind tibia, all near middle, the last one shorter and can be absent in some species. Besides this, the palpus is short and filiform, the upper calypter is glossiform and not reduced and the setae on sternite 5 are not bifurcated, as in *Palpilongus* gen. n.

*Cordiluroides* species can be recognized by the very high insertion of the antenna (very above the mid level of the eye), palpus short and slender, presence of only one pair of postsutural intra-alar seta, upper calypter transverse, hind tibia with one median anterodorsal, one posterior submedian and one posterodorsal supramedian setae and setae on sternite 5 not bifurcated. The genus was recently recorded from Costa Rica, on the base of three species (Couri et al. 2006).

## Palpilongus bifurcus sp. n.

http://zoobank.org/C1D0584D-A01B-4539-8A97-B60DCF53A209 http://species-id.net/wiki/Palpilongus\_bifurcus Figs 1–9

**Type material.** Holotype male: COSTA RICA: Prov. Guana [Guanacaste], Estation Pitilla 9 Km. S. de Santa Cecilia, 700m, DIC 1994, P. Rios, LN 329950 380450 #4372 [INBIO code collection number] (deposited at INBIO). Paratypes: Same locality as holotype, ix.1994, LN 330200\_380200 #3206, 1 female (INBIO), #3294, 1 female (INBIO); Prov. Alajuela, Upala, Bijagua, Alb. Heliconias, 700m, 11–26.i.2000, J. D. Gutiérrez, Agua miel, L\_N\_299800\_43800 #56263, 1 female (MNRJ), #56263, 1 female (INBIO); Prov. Guanacaste, Rio San Lorenzo, Tierras Morenas, 1050m, x.1995, G. Rodriguez, L\_N\_287800\_427600 #6405, 1 female (DZUP), # 6405, 1 female (INBIO); Prov. Punta [Puntarenas], Est La Casona, R.B. Monteverde, A.C. Arenal, 1520m, i.1994, N.G. Obando, LN 253250\_449700 #2606, 1 female (INBIO); N. P.



Figure 2. Palpilongus bifurcus gen. n. et sp. n. (holotype), mouthparts and palpus in lateral view.

Heredia Prov., Transecto, Braulio Carrillo, x.1989, 1500, R. Aguillar & M. Zumbado, 1 female (INBIO).

**Description. Male**. Body length – 3.8 mm. Wing length – 4.0 mm.

*Head.* Dichoptic. Ground-color yellow. Eye bare. Fronto-orbital plate, parafacial, face and gena golden pruinose. Frons brown-reddish about 1/3 of head width. Three pairs of frontal setae intercalated with shorter setae, the upper frontal setae oriented backwards. Ocellar setae strong. Antenna with pedicel yellow and postpedicel brown-ish, about 3.8 times the length of the pedicel. Arista with very short setulae. Gena thin. Palpus yellow, very long, as long as proboscis, enlarged toward apex. Labellum not reduced, developed and without teeth.

Thorax. Color yellow, with no marks. Acrostichal setae in front of suture in 3 irregular rows and after the suture in 1-2 rows, with some scattered cilia close to scutellar suture; notum with very few scattered cilia that increase somewhat in number on the scutellum; dorsocentrals 1+3, all long; intra-alars 1+2, supra-alars 2; postpronotals 2; prealar absent. Notopleuron without covering cilia and with two setae similar in size. Anepisternum with one row of six setae, one cilium on upper anterior margin and few scattered cilia on upper half. Prosternum bare, proepisternal depression bare, proepisternal setae oriented downwards. Katepisternals 1+1+1 and with scattered setulae. Meron and katepimeron bare. Scutellum with 1 short sub-basal seta, one lateral long and one apical a little shorter than the lateral one. Legs yellow. Fore femur with rows of posterodorsal and posteroventral setae. Fore tibia with 1 long median posterior seta, one strong dorsal seta on apical fourth, a pre-apical posterodorsal seta and an apical posteroventral seta. Tarsus with a long apical seta at base. Mid femur with short and thin row of scattered setae, ending with a longer seta; two pre-apical posterior setae. Mid tibia with 4 long dorsal setae on apical half (Fig. 3), one long and strong anterodorsal seta on apical third and long apical setae on each, the anterior, anteroventral and ventral, the last of which the longest and strongest. Hind femur with 2-3 fine anterodorsal and posterodorsal setae on apical third and with 3 preapical setae (anterodorsal, dorsal and posterodorsal). Hind tibia

30



Figure 3. Palpilongus bifurcus gen. n. et sp. n. (holotype), mid tibia.

with many series of fine and long anterodorsal, dorsal and posterodorsal setae and with a long and strong apical ventral seta. Wing slightly infuscated. Vein M straight. Calypters yellow. Knob of halter yellow.

*Abdomen.* Elongate-cylindrical. Ground-color yellow with black round lateral marks on tergites 3–5. Sternite 1 bare. Sternite 5 with setae on apical third, the marginal ones bifurcated (Fig. 4).

*Terminalia*. Cercal plate longer than wider, with setae on all surfaces and two small lateral spines on posterior margin (Figs 5–6); hypandrium tubular and phallic complex structures as in Fig. 5.

**Female**. Length of body: 4.5–5.3 mm. Length of wing, 4.8–5.5 mm. Differs from male as follows: proboscis with reduced labellum and strong teeth (Fig. 7). Mid femur has a median anterior seta. Mid tibia with one seta anterior to anterodorsal sub-basal seta and one supramedian posterior seta; hind femur with an anterodorsal and an anteroventral row of scattered setae and two long and thin ventral setae on the middle third. Hind tibia without the long setae as in male and with two anterodorsal setae at the limit of the median 1/3 and one submedian posterodorsal seta.

**Ovipositor.** Long, tergites 6–8 long and thin, covered with microtrichia, sternites 6–8 undivided, hypoproct long, cerci long (Figs 8–9). Three round spermathecae.

Distribution. Known only from Costa Rica.

**Remarks.** The male and the female have some marked differences, mostly in mid and hind leg chaetotaxy, in which only the male has mid tibia with four long dorsal

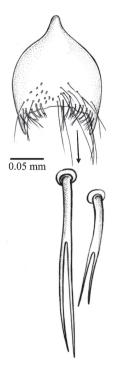
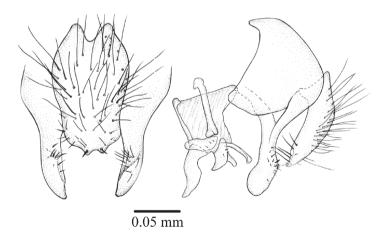


Figure 4. Palpilongus bifurcus gen. n. et sp. n. (holotype), sternite 5 with bifurcate setae in detail.

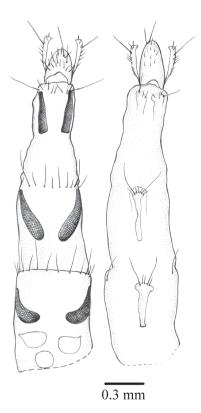


Figures 5-6. *Palpilongus bifurcus* gen. n. et sp. n. (holotype), cercal plate and surstyli 5 dorsal view 6 lateral view.

setae on apical half and hind tibia with many series of fine and long anterodorsal, dorsal and posterodorsal setae. The different shape of the proboscis suggests that feeding habits differs between males and females: the female certainly is predator as most spe-



Figure 7. Palpilongus bifurcus gen. n. et sp. n. (female paratype), head in lateral view.



Figures 8–9. *Palpilongus bifurcus* gen. n. et sp. n. (female paratype), ovipositor 8 dorsal view and spermathecae 9 ventral view.

cies of Coenosiini with reduced labellum and developed teeth, while the male posses another kind of habit, unknown as far we know.

**Etymology.** The specific epithet is Latin and refers to the bifurcate setae of sternite 5 of the male.

## Acknowledgments

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#### References

- de Carvalho CJB, Couri MS (2002) Part I. Basal groups. In: de Carvalho CJB (Ed) Muscidae (Diptera) of the Neotropical Region: Taxonomy. Editora Universidade Federal do Paraná, Curitiba, 1–132.
- de Carvalho CJB, Couri MS, Pont AC, Pamplona D, Lopes SM (2005) A catalogue of the Muscidae (Diptera) of the Neotropical Region. Zootaxa 860: 1–282.
- Couri MS, de Carvalho CJB (2002) Part II. Apical groups. In: de Carvalho CJB (Ed) Muscidae (Diptera) of the Neotropical Region: Taxonomy. Editora Universidade Federal do Paraná, Curitiba, 133–262.
- Couri MS, de Carvalho CJB, Pont AC (2006) *Cordiluroides* Albuquerque from Costa Rica: first records, descriptions and taxonomic changes (Diptera, Muscidae, Coenosiinae). Revista Brasileira de Entomologia 50: 341–346. doi: 10.1590/S0085-56262006000300003
- Couri MS, Pont AC (2000) Cladistic analysis of Coenosiini (Diptera: Muscidae: Coenosiinae). Systematic Entomology 25: 373–392. doi: 10.1046/j.1365-3113.2000.00125.x
- Couri MS, Salas C (2010) First record of *Coenosia attenuata* Stein (Diptera, Muscidae) from Chile with biological notes. Revista Brasileira de Entomologia 54: 144–145.
- Malloch JR (1934) Muscidae. In: Diptera de Patagonia e South Chile. London, Part 7: 171–346. doi: 10.1590/S0085-56262010000100020
- McAlpine JF (1981) Morphology and terminology adults. In: McAlpine JF, Peterson BV, Shewell GE, Teskey HJ, Vockeroth JR, Wood DM (Eds) Manual of Nearctic Diptera, volume 1. Research Branch, Agriculture Canada, Monograph 27, 9–63.
- Savage J, Vockeroth JR (2010) Muscidae (house flies, stable flies). In: Brown BV, Borkent A, Cumming JM, Wood DM, Woodley NE, Zumbado MA (Eds) Manual of Central American Diptera: Volume 2. NRC Research Press, Ottawa, Ontario, Canada, 1281–1295.
- Stuckenberg BR (1999) Antennal evolution in the Brachycera (Diptera), with a reassessment of terminology relating to the flagellum. Studia dipterologica 6: 33–48.

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RESEARCH ARTICLE



# Ancyronyx Erichson, 1847 (Coleoptera, Elmidae) from Mindoro, Philippines, with description of the larvae and two new species using DNA sequences for the assignment of the developmental stages

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#### Abstract

Ancyronyx buhid **sp. n.** and A. tamaraw **sp. n.** are described based on adults and larvae, matched using their cox1 or cob DNA sequence data. Additional records of A. schillhammeri Jäch, 1994 and A. minerva Freitag & Jäch, 2007 from Mindoro are listed. The previously unknown larva of A. schillhammeri is also described here, aided by cox1 data. The new species and larval stages are described in detail and illustrated by SEM and stacked microscopic images. Keys to the adult and larval Ancyronyx species of Mindoro and an updated checklist of Philippine Ancyronyx species are provided. The usefulness as bioindicators, the phylogenetic relationships and biogeographic aspects affecting the distribution patterns are briefly discussed.

## Keywords

*Ancyronyx*, Spider Water Beetles, Elmidae, Coleoptera, taxonomy, new species, larva, DNA barcoding, *cox1, cob*, Buhid, Mindoro, Philippines

#### Introduction

The so-called spider water beetles of the genus *Ancyronyx* Erichson, 1847, family Elmidae Curtis, 1830 are known from North America, China, South and Southeast Asia (Freitag 2012). The genus diagnosis was recently revised (Freitag 2012) due to the description of several new species in the last decade.

The genus appears closely related to Podelmis Hinton, 1941 but can be distinguish by the complete absence of a prosternal anterior process, and the shape of the terminal segment of the ovipositor (stylus slender and more or less straight versus somewhat conical and distinctly bent laterad in Podelmis). Two morphologically distinct groups of Ancyronyx are recognised, both are characterised by varying morphological and ecological adaptation patterns. The SE Asian representatives of the A. variegatus group (sensu Freitag and Jäch 2007) of slightly larger species can be found in mesosaprobic rivers. The relatively smaller species of the A. patrolus group (sensu Freitag and Jäch 2007) occur predominantly in clean permanent streams (Freitag 2012). When identified on species level, they might therefore serve as good bioindicators, just as Elmidae in general (e.g. Moog and Jäch 2003, Hilsenhoff 1982). As larvae of the respective taxa are naturally more abundant then adults, appropriate identification tools for these developmental stages might be of great interest for ecological and applied limnological studies. Larval stages of Ancyronyx have been formally described only from the Philippines so far (Freitag and Balke 2011), but the larval habitus of the only North American species was figured by Brown (1972).

This study is part of the Baroc River Catchment Survey of the Ateneo de Manila University which focuses on the Key Biodiversity Area "69 Hinunduang Mt." (*sensu* Ong et al. 2002), classified as a terrestrial and inland water area of very high biological importance and extremely high critical conservation priority ("EHc"), under high socioeconomic pressure (Ong et al. 2002), which is, however, only subjected to moderate conservation efforts and not yet formally protected (Ambal et al. 2012).

Furthermore, previously collected specimens from Mindoro were included, that are partly from other conservation and research priority areas of high and highest urgency ("64 Naujan Lake National Park" and "62 Puerto Galera" *sensu* Ong et al. 2002).

#### Material and methods

#### **Taxon Sampling**

The material was preserved in 95% ethyl alcohol to allow genetic sequencing. Most material was retrieved during the ongoing Baroc River Catchment Survey. Material collections from the 1990s were examined at the Naturhistorisches Museum Wien, Austria (NMW), the Senckenberg Museum für Tierkunde Dresden, Germany (SMTD), and the Zoological Museum of the University Copenhagen, Denmark (ZMUC). All specimens recorded by the first author were manually collected as indicated by letter "M" at the end of a collection label. Letter codes in parenthesis refer to a particular sampling station and microhabitat of the Baroc River Catchment. Number codes are arbitrary. They do not follow temporal or spatial patterns.

### DNA extraction and sequencing

DNA was extracted from five larvae and three adults (entire specimens) from Mindoro, and one entire adult specimen of the recently described Ancyronyx jaechi Freitag, 2012 from Sri Lanka using Qiagen DNeasy kit (Qiagen, Hilden, Germany). The extraction was done by a single elution following the protocol for animal tissues (Qiagen 2002). The 3' end of the cytochrome c oxidase subunit I (cox1) gene was amplified using polymerase chain reaction (PCR) following standard protocols (see http://zsm-entomology.de/wiki/ The\_Beetle\_D\_N\_A\_Lab) and using primer pairs C1-J-2183 (5'-CAA CAT TTA TTT TGA TTT TTT GG-3'; Jerry) and TL2-N-3014 (5'-TCC AAT GCA CTA ATC TGC CAT ATT A-3'; Pat) (Simon et al. 1994) and Mango Taq DNA polymerase (Bioline, Luckenwalde, Germany). The PCR temperature progression was set: 30 s at 94 °C, 30 s at 47 °C, 60 s at 72 °C (× 35 cycles), 600 s at 72 °C. Amplification products were purified with Qiagen Qiaquick PCR purification columns (Qiagen, Hilden, Germany). Cycle sequencing was performed as follows: 15 s at 96 °C, 15 s at 50 °C, and 240 s at 60 °C (x 35 cycles) using PCR primers with BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, California, USA). The sequencing products were purified by ethanol precipitation (25 µl of cold (-20°C) 99% ethanol, 2.5 µl of 3M sodium acetate added to product; centrifuged; washed with 25 µl of 70% ethanol), and additionally with Agencourt CleanSEQ (Agencourt Bioscience, Beverly, Massachusetts, USA) following protocol 000600v32 (Agencourt Bioscience 2006) before electrophoresis.

The DNA extraction of three specimens was additionally used for the amplification a central part of the cytochrome b apoenzyme (*cob*) gene by using the primer pair 5'-GAG GAG CAA CTG TAA TTA CTA A-3' (CB3) and 5'-AAA AGA AA(AG) TAT CAT TCA GGT TGA AT-3' (CB4) (Baraclough et al. 1999). This was done to prove assignment of adult and larval stages of one species for which *cox1* data were insufficient.

# **Phylogenetic analysis**

Additional *cox1* sequences of Philippine *Ancyronyx* species (Freitag and Balke 2011) previously submitted to ENA/GenBank (http://www.ebi.ac.uk/ena/) were included (see Table 1). The same applies for *Podelmis viridiaenea* Jäch, 1982 (Elmidae) from Sri Lanka that was used as outgroup. The newly amplified sequences were traced and aligned in CLUSTALW (Thompson et al. 1994) using BIOEDIT version 7.0.5.2. (Hall 1999) and default parameters. Phylogenetic analyses were conducted with MRBAYES vers. 3.1.2 (Ronquist et al.

2012) using the GTR (General Time Reversible) model (Tavaré 1986) with default priors starting with random trees with three heated and one cold Markov chains. The analysis was run by 1,000,000 generations, and the first 25% of samples from the cold chain have been discarded as burnin. Branch support for the Bayesian trees was assessed with posterior probabilities determined via the 50% majority rule consensus. This easy analysis is only intended for matching larva and adults of the species treated in this paper.

# Morphological analysis

Digital photographs were taken with an OLYMPUS SZ 61 stereo microscope (species habitus), and an OLYMPUS CX 21 compound microscope (dissected body parts), both with digital photo adapter LW Scientific MiniVID DCM 310. For each illustration a series of photographs taken at various focus layers was stacked using the stack function (species habitus) and corrected weighted average function (dissected body parts) of COMBINEZM software (Hadley 2008). The same optical systems we used for the dissection of adult specimens and the material examination. Biometric measurements were done by the use of a calibrated ocular micrometer.

Scanning electron microscope (SEM) images of vacuum dried material were obtained using a ZEISS EVO 50 XVP. Except for the single larval specimen of *Ancyronyx schillhammeri*, all specimens were coated with gold using one dissected and one entire specimen each.

For all larval material examined, measurements of the head capsule width are given in mm (e.g. 1 L (0.31)) as a suitable indicator for the larval size and the instar stage assignment (see Freitag and Balke 2011).

Morphological terminology follows Kodada and Jäch (2005) and Freitag and Balke (2011).

### Abbreviations and repositories

Brgy.	Barangay (local government unit district)
CL	calculated length (PL + EL)
EL	elytral length
EW	elytral width
HW	head width
ID	interocular distance
L	larva / larvae
Μ	manual collection
MW	maximum pronotal width
Oc.	Occidental
Or.	Oriental
PHIL	Philippines
PL	pronotal length

sec.veget.	surrounded by secondary vegetation
subm.	submerged
CFM	Collection Hendrik Freitag, Manila, Philippines, currently deposited at
	Ateneo de Manila University, Philippines
CZW	Collection Herbert & Salvacion V. Zettel, Vienna, Austria
NMW	Natural History Museum Vienna, Austria
PNM	Philippine National Museum Manila, Philippines
SMTD	Senckenberg Museum of Zoology Dresden, Germany
ZMUC	Zoological Museum of the University Copenhagen, Denmark
ZSM	Zoological State Collections Munich, Germany

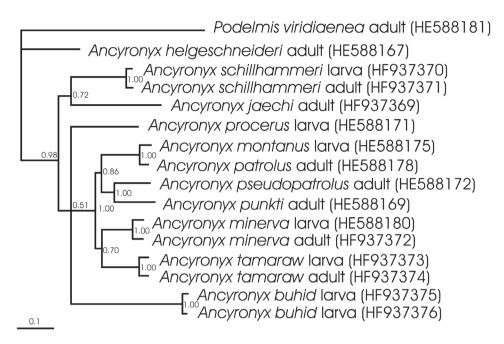
# **Data resources**

The data underpinning the analysis reported in this paper are deposited at GBIF, the Global Biodiversity Information Facility, http://ipt.pensoft.net/ipt/resource. do?r=ancyronyx\_mindoro\_data

All DNA sequences were submitted to ENA/GenBank via online submission to EMBL-EBI. Accession numbers and curatory information are listed in Table 1.

Species	Stage	Locality	Site	Voucher	cox1	cob
Ancyronyx jaechi Freitag, 2012	adult	Sri Lanka	1	ZSM FR 027	HF937369	-
Ancyronyx schillhammeri Jäch, 1994	adult	Mindoro	303a	ZSM FR 029	HF937371	-
Ancyronyx schillhammeri Jäch, 1994	larva	Mindoro	303a	ZSM FR 030	HF937370	-
Ancyronyx tamaraw Freitag	adult	Mindoro	302	ZSM FR 011	HF937374	-
Ancyronyx tamaraw Freitag	larva	Mindoro	302	ZSM FR 012	HF937373	-
Ancyronyx buhid Freitag	adult	Mindoro	318	ZSM FR 088	-	HF937366
Ancyronyx buhid Freitag	larva	Mindoro	HR2g	ZSM FR 090	HF937375	HF937367
Ancyronyx buhid Freitag	larva	Mindoro	310a	ZSM FR 091	HF937376	HF937368
Ancyronyx minerva Freitag & Jäch, 2007	adult	Mindoro	303a	ZSM FR 033	HF937372	-
Ancyronyx minerva Freitag & Jäch, 2007	larva	Palawan	159	ZSM FR 025	HE588180	-
Ancyronyx helgeschneideri Freitag & Jäch, 2007	adult	Palawan	CR4	ZSM FR 007	HE588167	HE588183
Ancyronyx procerus Jäch, 1994	larva	Busuanga	169	ZSM FR 014	HE588171	HE588182
Ancyronyx montanus Freitag & Balke, 2011	larva	Palawan	16h	ZSM FR 038	HE588175	-
<i>Ancyronyx punkti</i> Freitag & Jäch, 2007	adult	Palawan	154	ZSM FR 008	HE588169	-
Ancyronyx pseudopatrolus Freitag & Jäch, 2007	adult	Palawan	16f	ZSM FR 003	HE588172	-
Ancyronyx patrolus Freitag & Jäch, 2007	adult	Busuanga	165	ZSM FR 032	HE588178	-
Podelmis viridiaenea Jäch, 1982	adult	Sri Lanka	1	ZSM FR 035	HE588181	-

**Table 1.** ENA/GenBank accession numbers of DNA sequences, geographical origins, collection sites and organismic sample references of specimens used for molecular-genetic analyses.



**Figure 1.** Phylogram of the consensus tree of the Bayesian analysis with branch lengths measured in expected substitutions per site. Posterior probability values (printed when > 0.5) at respective branches. Sample labels with developmental stage and ENA/GenBank code.

# Results

### **DNA** sequence analysis

Alignment of the *cox1* data and trimming ambiguous bases at the 3' and 5' ends yielded a matrix of 804 bp. None of the sequences contained indels. The sequences of the larvae of *A. buhid* had nine ambiguous positions in-between which were coded as 'N's.

All adults and larvae could be matched unambiguously. Sequences of adult and larva of the same species from the same locality or island were identical or varied just in one base pair. Sequence samples of *A. minerva* from Mindoro diverged in eight base pair positions (six of them synonymous substitutions) from that of the same species from Palawan.

The sequencing of *cox1* of the adults of *A. buhid* failed and is not included in the phylogenetic analysis. The *cob* sequences, however, which were amplified for two larvae and an adult of this species allowed unambiguous matching of the developmental stages. Their aligned and trimmed particial *cob* sequences of 350 bp were identical except for four positions where a synonymous substitution was seen in one of either sequences.

A 50% majority rule consensus trees based on *cox1* data is illustrated in Fig. 1. All samples of the same species clustered together, supported by 1.0 posterior probability values. The species of the *A. patrolus* species group and its two subgroups respectively clustered together, however partly with lower posterior probability value support. *A. buhid* does not cluster with the *A. patrolus* group.

# Taxonomy

# Ancyronyx minerva Freitag & Jäch, 2007

http://species-id.net/wiki/Ancyronyx minerva\according to Freitag 2013 Figs 2, 5

Ancyronyx minerva Freitag & Jäch, 2007: 50–53 (adult description); Freitag and Pangantihon 2010: 133 (first record Mindoro); Freitag and Balke 2011: 53–58 (larva description), 79 (key); Freitag 2012: 63 (world check list).

**Material examined.** 1  $\bigcirc$ , 6 L (0.22; 0.25, 2 × 0.29, 0.31) (CFM) "PHIL.: Mindoro, Puerto Galera, NR km 37.2, downstr. Tamaraw Falls; riffle&fall; rocks,woodlitter,roots; sec.veget.;c.80m asl., 13°27'03"N 120°59'27"E 22.4.1994, leg. Freitag (302)M"; 1 Q [FR015], 1 🖉 [FR033], 1 L (0.31) (ZSM) "PHIL.: Mindoro, San Teodoro, Tukuran Riv.; small lowld.riv.;riffle &run; woodlitter, gravel; sec.veget.; c.30m asl., 13°25'34"N 120°58'37"E 23.4.1994, leg. Freitag (303a) M";1 & (CFM) "PHIL.: Mindoro, Puerto Galera, NR km 59, downstr. Aninuan Falls; riffle; small mount. riv.,boulder,rocks,gravel,woodlitter; sec.veget.;c.80m asl., 13°29'10"N 120°54'18"E 24.4.2009, leg. Freitag (304)M"; 1 ♀ pter., 2 L (0.21, 0.25 [FR079, FR080]) (ZSM) "PHIL.: Mindoro Oriental, Municipality Victoria, Brgy. Malayas, Malayas River; W Naujan tributary; sec. veget., submerged wood, riffle, c.20 m asl., c.13°09'26"N 121°18'29"E; 22.2.2010 leg. Freitag & Pangantihon (308a)M"; 1 d pter. (CFM) "PHIL.: Mindoro Oriental, Bongabong, Brgy. Formon, Pastuhan, Tangisan Falls; deep mountain valley, sec. forest, submerged wood, riffle, c.200 m asl., c.12°43'N 121°23'E; 27.10.2011 leg. Freitag (318a)M"; 1 Q, 2 L (0.24 [FR087], 0.27) (ZSM, CFM) "PHIL.: Mindoro Oriental, Bongabong, Brgy. Formon, Pastuhan, Tangisan Falls; deep mountain valley, sec. forest, gravel & boulders, riffle, c.200 m asl., c.12°43'N 121°23'E; 27.10.2011 leg. Freitag (318)M"; 1 Q (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Baroc River; subm. wood; gravel flood plains; c.12°37'07"N 121°24'06"E, 90m asl; leg. Freitag 1 Apr.2013(BRf)M"; 1 ♂, 1 ♀ (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente proper, Taugad River; subm. wood; sec.veget.; c.12°37'06"N 121°23'49"E, 100m asl; leg. Freitag 2 Apr.2013(TR1f)M"; 1 Å, 1 ♀ 3 L (0.29, 2 × 0.31) (CFM) "PHIL: Or.Mindoro, Roxas, Brgy. San Vicente, Tauga River; rocks, riffle & run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Freitag 17.11.2011(TR2g) M"; 1♂, 1♀, 3 L (0.27, 0.29, 0.31) (CFM): same locality and microhabitat "leg. Freitag & Pangantihon 07.7.2012 (TR2g)M"; 1 ♀ (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Baroc River tributary Hiyong Creek; bottom gravel, run & riffles; sec.veget.; c.12°37'27"N 121°22'48"E, 147m asl; leg. Pangantihon, 29.Jun.2012(THCc)M"; 2 L (0.24, 0.25) (CFM) "PHIL:Or. Mindoro, Roxas, Brgy. San Vicente, Baroc River tributary Hiyong Creek; subm. root packs, run; sec.veget.; c.12°37'27"N 121°22'48"E, 147m asl; leg. Freitag & Pangantihon, 07.Jul.2012(THCh)M"; 1 L (0.31) (CFM) "PHIL: Or. Mindoro, Roxas, Brgy.



Figures 2-9. Habitus (not to scale) of 2 Ancyronyx minerva, adult 3 A. tamaraw, sp. n. adult 4 A. buhid, sp. n., adult 5 A. minerva, larva 6 A. tamaraw, sp. n., larva 7 A. buhid, sp. n., larva 8 A. schillhammeri, adult 9 A. schillhammeri, larva.

San Vicente, Sitio Quirao, Hinundugan tributary Quirao Buhay Creek; rocks, run; 12°36'10"N 121°23'00"E, 130m asl; leg. Freitag & Pangantihon, 30.06.2012 (HBCg)M".

**Distribution.** The species is known from Busuanga, Mindoro and Palawan (Philippines) and is common on these islands.

**Remarks.** Morphological variations between the population of different islands are not evident, but the *cox1* gene varies in a few more substitutional sites than within a population of one and the same island. The larva was described and illustrated in Freitag and Balke (2011).

**Ecology.** Both, adults and larvae, are usually collected from boulder and rock surfaces, or submerged rootpacks in run and riffle sections. The species is predominantly found in clean, small to medium sized permanent streams. (Freitag and Pangantihon 2010, Freitag and Jäch 2007).

#### Ancyronyx tamaraw Freitag, sp. n.

http://zoobank.org/9D03899B-56E5-4E7C-9C0E-A5154E30B2FC http://species-id.net/wiki/Ancyronyx\_tamaraw Figs 3, 6, 10A–M, 11A–I, 12A–H

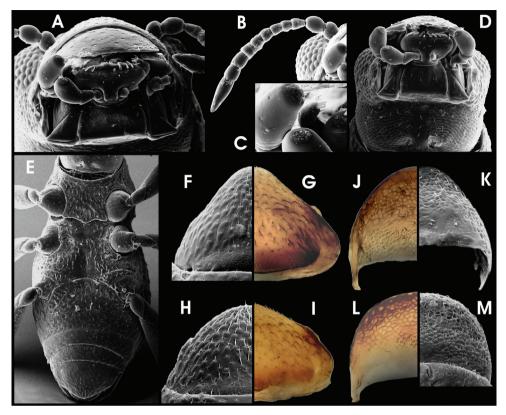
**Etymology.** This small and probably rare species is named in reference to its type locality, the Tamaraw Falls on the island of Mindoro. The tamaraw is a small Mindoro-endemic buffalo. The term is used as noun in apposition.

**Type material. Holotype**  $\mathcal{F}$  (NMW) "PHIL: Mindoro, Puerto Galera, NR km37.2 downstr. Tamaraw Falls; riffle&fall; rocks, woodlitter, roots; sec.veget.; c.80m asl., 13°27'03"N 120°59'27"E 22.4.1994, leg. Freitag (302)M", terminal parts of abdomen incl. aedeagus glued separately. **Paratypes:** 18  $\mathcal{QQ}$ , 15  $\mathcal{F}$  (NMW, ZSM [FR011], ZMUC, SMTD, CFM), 9 L (0.24, 0.28, 5 × 0.29, 2 × 0.30) (ZSM [FR012], NMW, SMTD, CFM): same label data as holotype. 3  $\mathcal{QQ}$ , 4  $\mathcal{F}$  (PNM): same locality data as holotype, leg. Freitag 11.2.2013.

Adult description. Body 1.2–1.5 mm long (CL + exposed portions of head & tergit VIII); CL: 1.02–1.36 mm; EW: 0.51–0.58 mm, CL/EW: 2.0–2.4.

Colouration as in Fig. 3: ventral side, coxae, trochanter, and pronotum brown; entire dorsal head capsule and mouthparts dark brown; elytra dark brown except for two pairs of yellow patches; anterior yellow elytral patches round, extending each between first and third row of elytral punctures, not reaching median or anterior elytral margin; posterior yellow elytral patches elongate-oval to subtriangular, not reaching median, lateral, and apical elytral margin; antennae yellow (except for dark tips and scape); legs dominantly yellowish except for coxa, trochanter and brown areas around all articulations, especially proximal and distal areas of femur, proximal third of tibia and distal third of fifth tarsomere.

HW 0.29–0.35 mm; ID 0.15–0.18 mm; labrum and distal portion of clypeus moderately densely micropunctate and covered with short trichoid setae (Fig. 10A); proximal portion of clypeus and frons microreticulate and punctate; frontoclypeal suture inconspicuous, slightly convex. Eyes slightly protruding. Antennae (Fig. 10B) with 11 antennomeres, slender, c. as long as head wide. Genae (Fig. 10D) rugose and



**Figure 10.** *Ancyronyx tamaraw* Freitag, sp. n., (SEM photographs in greyscale, stereo microscope photographs in colour; not to scale: see respective chapter for size measurements); adult male: **A** head, frontal **B** antenna, frontal **C** maxillary and labial palpi, frontal **D** head, ventral **E** throax and abdomen, ventral; adult female: **F**, **G** ventrite 5, ventral; adult male: **H**, **I** ventrite 5, ventral; adult female: **J**, **K** tergite VIII, dorsal; adult male: **L**, **M** tergite VIII, dorsal.

reticulate, with indistinct pubescence. Gula (Fig. 10D) with somewhat regularly arranged striae except for central portion, moderately densely pubscent; gular sutures absent. Mandible with bilobed tip. Maxilla (Fig. 10A) with very short cardo; stipes with distinct flat, triangular elevation ventrally; galea palp-like elongate, with apical setae and sensilla (Fig. 10C); lacinia not examined. Labium (Fig.10A) with subtrapezoidal postmentum, prementum suboval, undivided, with subapical row of ten short trichoid setae; labial palps three-segmented, c. as long as postmentum, with apical setae and sensilla (Fig. 10C).

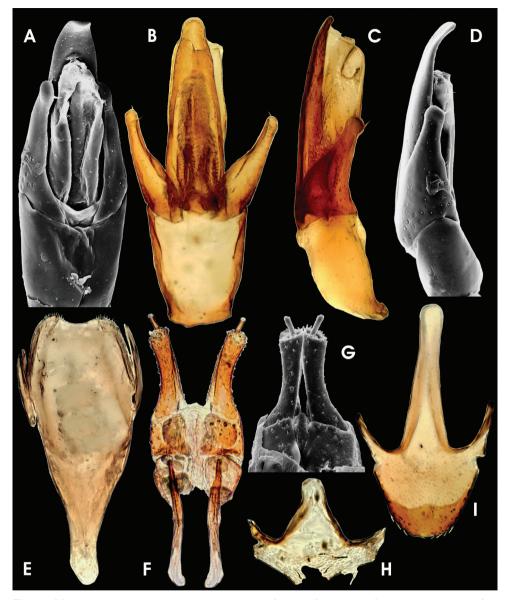
Pronotum (Fig. 3) 0.35–0.38 mm long (PL), 0.38–0.41 mm wide (MW), inconspicuously wider than long (PL/MW), widest at about posterior 0.4, distinctly narrower than elytra, with moderately deep transverse groove; anteriorly of transverse groove slightly vaulted; posterior portion medially elevated; posterolateral oblique grooves small and round, but conspicuous; lateral margin distinctly arcuate; anterior margin convex; pronotal surface entirely microreticulate and with moderately densely arranged seta-bearing tubercles; lateral pronotal carina absent; hypomeron inconspicuously reticulate. Prosternum (Fig. 10E) punctate; prosternal process broadly subpentagonal, distinctly wider than long, almost flat.

Metascutellum subcordiform, medially slightly impressed, micropunctate. Elytra (Fig. 3) elongate, 0.78–0.98 mm long (EL), c. 1.5–1.7 times as long as wide (EL/EW), laterally arcuate (broadest at about anterior 0.45), anteriorly slightly roundly convergent, posteriorly roundly convergent to apices, with eight longitudinal, moderately impressed rows of punctures (counted at level of metacoxae); median rows more regular than lateral ones; five strial rows between suture and humerus; punctures large and moderately deeply impressed; interstices and intervals convex, granulose to microreticulate; lateral elytral gutter very narrow, inconspicuous; humeri broadly rounded; elytral apices separately rounded. Mesoventrite (Fig. 10E) short, micropunctuate, with a round median impression and a sublateral pair of round elevations. Metaventrite (Fig. 10E) comparably small; medial impression wide, not conspicuously longitudinal, rather a shallow, funnel-like round impression deepest at median posterior margin; disc with scattered inconspicuous setose tubercles, glabrous in-between. Anepisternum 3 microreticulate with additional scattered punctures. No hind wings present in all specimens examined.

Legs (Fig. 3) slightly longer than body; coxae large; pro- and mesocoxae (Fig. 10E) subglobular (drop-shaped), lateral portion visible in dorsal view; metacoxae (Fig. 10E) rather conically elevated from a flat base, not visible in dorsal view; trochanter (Fig. 10E) small, broadly lanceolate, invisible in dorsal view; femora and tibiae with microsetiferous tubercles; tibiae distally with few trichoid setae; each tarsomere with ventral pair of short trichoid setae; claws moderately wide, rather short (compared to other *Ancyronyx* species), strongly bent, base of each with two teeth, distal one distinctly larger.

Ventrite 1 (Fig. 10E) distinctly arcuately projected anteriad; medioanterior portion depressed (connecting to funnel-like metaventrite impression). Ventrites 2–4 (Fig. 10E) with small, moderately densely arranged punctures; surface between punctures glabrous; tubercles larger and denser toward lateral declivity; ventrite 5 (Figs 10F–I) moderately densely covered with short adpressed setae emerging from flat tubercles; lateral projection inconspicuous.

Aedeagus (Figs 11A–D) 410  $\mu$ m long, somewhat similar to that of *A. sophiemarie* Jäch, 2004 (see Jäch 2004: figs 3–4), but phallobase longer and apical area of median lobe distinctly wider. Phallobase almost symmetrical, more or less straight, except for tapered and ventrally bent base, slightly longer ventrally, with conspicuous, strongly sclerotised ventral and lateral margins. Median lobe moderately long and wide (c. 70  $\mu$ m), straight, evenly and slightly tapering towards apex up to apical 0.2 of median lobe, then moderately bent ventrad and more abruptly tapering into a wide apical area; apex pointing ventrad, with numerous pore-like structures on dorsal side; basolateral (penile) apophyses short, not overreaching paramere base; ventral sac distinct, ventrally protruding (Fig. 11C), internal portion speckled, moderately sclerotised (Fig. 11B). Fibula weakly sclerotised; corona inconspicuous. Parameres short, c. 145  $\mu$ m long, reaching about basal 0.7 of aedeagus, elongately subtriangular, widely separated



**Figure 11.** *Ancyronyx tamaraw* Freitag, sp. n., (SEM photographs in greyscale, stereo microscope photographs in colour; not to scale: see respective chapter for size measurements); adult male: **A** aedeagus, ventral **B** aedeagus, dorsal **C**, **D** aedeagus, lateral **E** sternite IX, ventral; adult female: **F**, **G** ovipositor, ventral; adult male: **H** sternite VIII, ventral; adult female: **I** sternite VIII, ventral.

ventrally; laterobasal margin emarginate (Fig. 11D) apical portion roundly widened, narrowest subapically, with few short setae (two apical, two medio-subapical).

Sternite IX (Fig. 11E), c. 380  $\mu$ m long, with short anterior strut, not clearly partitioned from apical portion; apical corners rounded, with patches of micro-setae, apical margin broadly emarginate; longer paraproct reaching apical margin.

Ovipositor (Fig. 11F, G) c. 480  $\mu$ m long. Stylus slender, rather staight, with various sensilla. Coxite long, outer margin curved, distal portion with several rather short and broad, peg-like spines, most densely set subapically at lateral margins; inner margin pubescent; basal portion with similar, slightly slenderer, peg-like spines; near valvifer insertation with dense patch of very small spines. Valvifer as long as coxite; fibula distinctly bent and widened caudally.

Secondary sexual characters: Sternite VIII in male (Fig. 11H) short, weakly sclerotised and with very short median strut; in female (Fig. 11I) distinctly longer, slightly more sclerotised than in male, apical corners broadly rounded and with small seate, median portion with dense micro-pubescence (not conspicuous in males). Tergite VIII in female (Figs 10J, K) long, subtriangular, slightly longer than broad (c. 220  $\mu$ m long, 210  $\mu$ m wide), with few moderately short setae (apical ones widest), condyles large and conspicuous. Tergite VIII in male (Figs 10L, M) subsemicircular, distinctly wider than long and shorter than in female (c. 180  $\mu$ m long, 225  $\mu$ m wide); in apical half with moderately long setae. Ventrite 5 in female (Figs 10F, G) subtriangular (c. 250  $\mu$ m long, 360  $\mu$ m wide); in male (Figs 10H, I) similar in general shape, but slightly shorter (c. 210  $\mu$ m long, 370  $\mu$ m wide) and rounder.

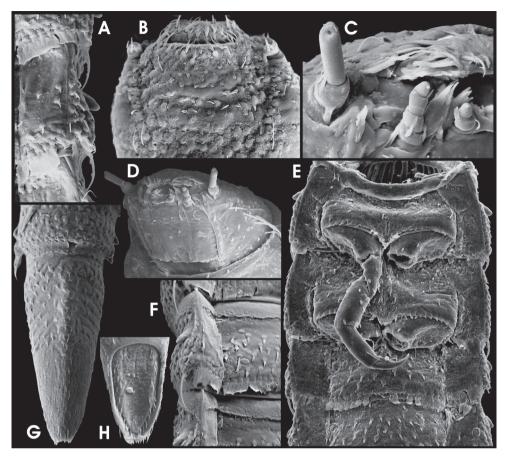
Adult differential diagnosis. Ancyronyx tamaraw superficially resembles A. sophiemarie from Sibuyan and A. minerva. The new species can be easily distinguished by the combination of elytral colour pattern (anterior yellow elytral patches circularly round, not reaching median or anterior elytral margin; posterior patches elongate-oval to subtriangular), the predominantly yellowish legs, the brown (not black) pronotum and head, and it's aedeagus with wide and flat apical portion.

Larval diagnosis (based on sixth instar). Colour (Fig. 6) similar to that of *Ancyronyx minerva* (see Freitag and Balke 2011: figs 3, 11A–L), but most distinctly different by anterior median head portions (clypeus, anterior frons) pale; anterior yellow pronotal band small, limited to very most anterior portion; at least pro-, meso-, metanotum with small circular-round (not broadly subtriangular) yellow pattern at medioposterior margin; abdominal segment IX with pale yellowish apex and a conspicuous dark pattern extending c. posterior 0.2–0.4; abdominal segment IX relatively longer than in larvae of *A. minerva*.

HW 0.29 mm; entire larva about 2.7 mm long. Body elongate very similar in the external characters to that of *A. minerva*, except for the following: Posterolateral projections (Figs 12A, F) of all abdominal segments short, generally not overreaching posterior segment margins.

Head (Figs 6, 12B–D) with subparallel lateral margins posterior 0.1–0.7, moderately tapering anteriad; lateral setae long; a dorsolateral pair of moderately long single setae present (Fig. 12B). Frontal suture distinctly V-shaped. Labrum subtrapezoidal. Antennae (Figs 12C, D) c. ¼ as long as head; peduncle at with at least one faciculate seta; pedicel c. two times as long as scape, c. three times as long as wide. Maxillary stipes (Fig. 12D) slightly tapering towards apex. Labial mentum (Fig. 12D) narrowest basal; lateroapical pair of spines rather small, positioned at distal edge.

Pro-, meso-, metathorax and legs (Figs 6, 12E) almost as in *A minerva*. Pronotum with rather inconspicuous small round signa (glabrous areas) in posterior half. Ventral sclerites of thorax (Fig. 12E) rugulose, not glabrous.



**Figure 12.** Ancyronyx tamaraw Freitag, sp. n., larva (SEM photographs; not to scale): **A** detail of abdominal segment III, lateral, with posterolateral projections and spiracles **B** head, dorsal **C** head, frontal **D** head, ventrolateral **E** meso- and metathoracic and first and second abdominal segments, ventral **F** lateral parts of median sclerites of abdominal venter, with setiferous tubercles and lateral projections **G** abdominal segments VIII and IX, dorsal **H** operculum, ventral.

Abdomen (Figs 6, 12A, E–H) without conspicuous dorsosagittal carinae except for the anterior half of segment IX (Fig. 12G); squamose setae at posterior rim of segments I–VIII large (Figs 12E, F). Ventral sclerites of segment I with distinct sagittal ridge in anterior half, reaching c. ½ to ⅔ of segment length. Apex of segment IX (Fig. 12G) emarginate (sometimes inconspicuous due to apical setae). Operculum (Fig. 12H) longer than in *A. minerva* (more than double as long as wide).

**Larval differential diagnosis.** The species can most easily be distinguished from *A. minerva* which looks superficially most similar by the partly pale colour pattern of the dorsal head, the narrower circular medioposterior pale pattern at pro-, meso-, and metanotum and the longer last abdominal segment with pale apical area and distinctly dark subapical portion.

**Variation between larval instars.** The final and prefinal instar stages available for this study do not vary conspicuously except for their size.

**Distribution.** The species is known from the type locality in north-eastern Oriental Mindoro. Additional material that appears conspecific is known from Subic, Zambales, Luzon and Bohol (unpublished material at CFM and NMW).

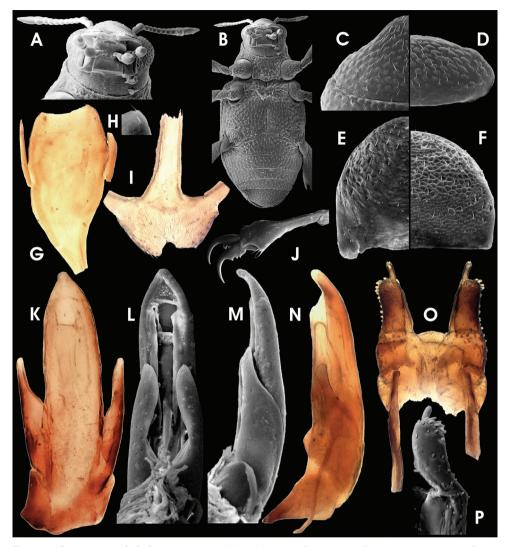
**Ecology.** The specimens were collected in well oxygenated water from rock surfaces, submerged woodlitter and roots in run and riffle sections of the stream below Tamaraw Falls. Since all examined material comes from this, in fact clean and almost natural small mountain river, any detailed conclusion about the habitat and ecological requirements would be highly speculative. It is, however, surprising that not any single specimen was found at any other collection site in Mindoro so far.

#### Ancyronyx bubid Freitag, sp. n.

http://zoobank.org/C731192C-EF06-4BDD-A387-9AB967DC9FED http://species-id.net/wiki/Ancyronyx\_buhid Figs 4, 7, 13A–P, 14A–I

**Etymology.** The species is named for the indigenous ethnic group of the Buhid in whose ancestral areas it commonly occurs. Same time, their kind support and care during regular field trips of faculty members and students of the Ateneo de Manila University's Biology Department to the their Ancestral Domain should be honoured. Buhid is used as noun in apposition.

**Type material. Holotype**  $\mathcal{J}$  (NMW) "leg. Jäch 1.12. \ PHILIPPINEN – Mindoro \ 20km W Calapan 1992 \ Hidden Paradise (21)", terminal parts of abdomen incl. aedeagus glued separately. **Paratypes**: 11  $\bigcirc \bigcirc$ , 23  $\bigcirc \bigcirc$ , 6 L (3 × 0.29, 2 × 0.30, (0.31) (NMW): same data as holotype. 8 L (2 × 0.29, 0.30, 4 × 0.31, 0.32) (NMW) "PHILIPPINEN – Mindoro \ 20km W Calapan 1992 \ Hidden Parad. 20.–21.11. \ leg. Jäch(10)"; 4 9 9 "PHILIPPINEN – Mindoro \ 20km W Calapan 1992 \ Hidden Parad. 20.–21.11. \ leg. Schillhammer (10)";  $3 \stackrel{\circ}{\bigcirc} \stackrel{\circ}{\bigcirc}, 3 \stackrel{\circ}{\subsetneq} \stackrel{\circ}{\ominus}$  (CZW) "PHIL.: Mindoro or. \ Baco, Hidden Parad. \ 19.-20.11.1993 \ leg. Zettel (27)"; 7 33, 4 99, 4 L (0.27[FR091]), 2 × 0.31, 0.29) (SMTD) "PHIL.: Mindoro, Baco, Dulangan, Lantuyan torrent mount. Riv.; sec.veget.; riffle, wood debris, c.55m asl., c.13°16'08"N 121°04'56"E 02.4.2000, leg. Freitag (310a)M"; 1 ♂, 3 ♀♀ (PNM, ZSM [FR088]) "PHIL.: Mindoro Oriental, Bongabong, Brgy. Formon, Pastuhan, Tangisan Falls; deep mountain valley, sec. forest, submerged wood, riffle, c.200 m asl., c.12°43'N 121°23'E; 27.10.2011 leg. Freitag (318)M"; 11 L (0.22, 0.24, 0.25, 6 × 0.31, 2 × 0.32) (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Tauga River; rocks, riffle & run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Freitag 17.3.2012(TR2g)M"; 1 👌 [FR086] (ZSM), 4 L (0.21, 0.24, 0.29, 0.31) (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Tauga River; rocks, riffle & run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Freitag & Pangantihon 07.7.2012(TR2g)M"; 3 ♂♂, 8 ♀♀ (SMTD) "PHIL:Or.Mind-



**Figure 13.** *Ancyronyx buhid* Freitag, sp. n., (SEM photographs in greyscale, stereo microscope photographs in colour; not to scale); adult male: **A** head, ventral **B** entire body, ventral **C** adult female: ventrite 5, ventral; adult male: **D** ventrite 5, ventral; adult female: **E** tergite VIII, dorsal; adult male: **F** tergite VIII, dorsal **G** sternite IX, ventral; aedeagus, ventral; adult female: **I** sternite VIII, ventral; adult male: **J** proximal tarsal segment, lateral **K** aedeagus, dorsal **L** aedeagus, ventral; **M**, **N** aedeagus, lateral **E** adult female: **O**, **P** ovipositor, ventral.

oro, Roxas, Brgy. San Vicente, Tauga River; subm. wood, run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Pangantihon, 22 Jan.2013(TR2f)M"; 2  $\bigcirc \bigcirc$ , 2  $\bigcirc \bigcirc$  (ZSM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Tauga Diit, Baroc River tributary Tauga Diit; subm. wood, run & riffle; sec.veget.; 12°37'32"N 121°21'17"E, 180m asl; leg. Freitag & Pangantihon, 05.Feb.2012(TIRf)M"; 2  $\bigcirc \bigcirc$  (CFM) "PHIL:Or. Mindoro, Roxas, Brgy. San Vicente, Baroc River tributary Hiyong Creek; side pool, lit-

toral sand and gravel; sec.veget.; c.12°37'27"N 121°22'48"E, 147m asl; leg. Freitag & Pangantihon, 05.Feb.2012(THCe)M"; 6 L (0.22, 2 × 0.24, 0.27, 2 × 0.31) (NMW) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Baroc River tributary Hiyong Creek; subm. root packs, run; sec.veget.; c.12°37'27"N 121°22'48"E, 147m asl; leg. Freitag & Pangantihon, 07.Jul.2012(THCh)M"; 3 33, 7 99, 3 L (0.27, 0.30, 0.31) (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Tauga Diit, Baroc River tributary Tauga Daka; subm. wood in run; sec.veget.; c. 12°37'56"N 121°20'33"E, c.350m asl; leg. Freitag, 4 Apr. 2013 (TDR2f)M"; 15 ♂♂, 14 ♀♀ (NMW) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Tauga Diit, Baroc River tributary Tauga Daka; subm. wood in run; sec.veget.; c. 12°38'05"N 121°19'33"E, c.530m asl; leg. Pangantihon, 18 Jan. 2013 (TDR3f)M"; 4 ♂♂, 7 ♀♀ (ZMUC) same locality and microhabitat "leg. Pangantihon, 23 Jan. 2013 (TDR3f)M"; 16 ♂♂, 14 ♀♀ (CFM) same locality and microhabitat "leg. Pangantihon, 15 Feb. 2013 (TDR3f)M"; 3 99, 2 L (0.27, 0.34) (PNM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Tagaskan, Hinundugan River,; rocks, run; sec.veget.; c.12°35'22"N 121°21'54"E, c.200m asl; leg. Freitag & Pangantihon,20.12.2011(HR2g)M"; 4L (0.25 [FR090], 0.26, 2 × 0.31, 0.32) (ZSM): same locality and microhabitat "leg. Freitag & Pangantihon,06.Feb.2012(HR2g)M";  $2 \, \text{dd}$ ,  $5 \, \text{QQ}$  (CFM) "PHIL: Or. Mindoro, Roxas, Brgy. San Vicente, Sitio Tagaskan, Hinundungan River; subm. wood, run c.12°36'30"N 121°22'38"E, c.200m asl; leg. Pangantihon; 31 Mar. 2013(HR3f)M"; 6 ♂♂, 1 ♀, 9 L (2 × 0.24, 2 × 0.27, 3 × 0.30, 2 × 0.33) (CFM) "PHIL: Or. Mindoro, Roxas, Brgy. San Vicente, Sitio Tagaskan, Hinundungan River; rocks, riffle & run; c.12°36'30"N 121°22'38"E, c.200m asl; leg. Freitag; 31 Mar. 2013(HR3g)M"; 3 L (0.24, 0.25, 0.31) (CFM) "PHIL: Or. Mindoro, Roxas, Brgy. San Vicente, Sitio Quirao, Hinundugan tributary Quirao Buhay Creek; rocks, run; 12°36'10"N 121°23'00"E, 130m asl; leg. Freitag & Pangantihon, 30.06.2012 (HBCg) M"; 1 L (0.30) (CFM) "PHIL: Or. Mindoro, Roxas, Brgy. San Vicente, Hinundugan tributary Tinggiwang Creek; subm. wood, run; 12°35'48"N 121°22'00"E, c.180m asl; leg. Freitag; 31 Mar.2013 (HTCf)M". Other material: 1 👌 (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Tauga River; subm. wood, run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Freitag 28.Nov.2011 (TR2f)M".

**Adult description.** Body 1.4–1.6 mm long (CL + exposed portions of head & tergit VIII); CL: 1.25–1.38 mm; CL/EW: 1.9–2.1. Colouration as in Fig. 4: entire dorsal head capsule, mouthparts, pronotum, and elytra (except for two pairs of yellow patches) black; anterior yellow elytral patches extending from humeri mediad approximately up to second row of elytral punctures, not reaching median elytral margin; posterior yellow elytral patches oval, not reaching median, lateral, and apical elytral margin; ventral side, coxae, trochanter, femur, proximal half of tibia, areas around tibial and tarsomere articulations, and claws brown; at least distal half of tibia and proximal portion of fifth tarsomere yellowish; antennae yellow (except for dark tips and basal segment).

Head (Figs 4, 13A) 0.33–0.36 mm wide (HW); ID 0.19–0.21 mm; labrum smooth, with moderately densely trichoid pubescence; clypeus (except for anterior margin) and frons with longitudinal striae on microreticulate ground, moderately

densely covered with short trichoid setae; frontoclypeal suture straight and conspicuous. Eyes slightly protruding. Antennae (Fig. 13A) with 11 antennomeres, slender, slightly shorter than head width. Genae (Fig. 13A) reticulate, with indistinct pubescence. Gula (Fig. 13A) with regularly arranged, scale-like striae (including median portion), with inconspicuous pubscens; gular sutures absent. Mouthparts (Fig. 13A) almost as in *A. tamaraw* except for shorter postmentum, that is rather sub-rectangular than trapezoidal; prementum with subapical row of eight very short trichoid setae; labial palps three-segmented, slightly longer then postmentum.

Pronotum (Fig. 7) 0.36–0.40 mm long (PL), 0.42–0.44 mm wide (MW), slightly wider than long (PL/MW), widest at about posterior 0.3, distinctly narrower than elytra, with deep transverse groove; anteriorly of transverse groove slightly vaulted; posterior portion broadly vaulted; posterolateral oblique grooves divided (two pairs), elongate, conspicuous; lateral margin distinctly arcuate; anterior margin distinctly convex; pronotal surface entirely microreticulate and rugose, with inconspicuous pubescence; lateral pronotal carina absent; hypomeron as pronotal surface. Prosternum (Fig. 13B) transverse, prosternal process broadly subpentagonal, distinctly wider than long, both appearing rugose by microreticulation superimposed with irregularly shaped setiferous tubercles.

Metascutellum subcordiform, micropunctate. Elytra (Fig. 4) broadly elongate, 0.89–0.98 mm long (EL), 0.61–0.66 mm wide (EW), c. 1.4–1.5 times as long as wide (EL/EW), almost parallel-sided in anterior 0.1–0.65, anteriorly slightly convergentposteriorly roundly convergent to apices , with c. nine longitudinal, moderately impressed rows of punctures (counted at level of metacoxae); median rows rather inconspicuous; lateral rows more regular and more deeply impressed than median rows; six to seven strial rows between suture and humerus; punctures moderately large and moderately deeply impressed, lateral punctures deeper than median ones; interstices and intervals granulose to micropunctate; lateral elytral gutter narrow; humeri roundly obtuse; elytral apices inconspicuously separately rounded.

Mesoventrite (Fig. 13B) very short, most anteriorly micropunctuate, posteriorly granulose, with deeply impressed median longitudinal impression. Metaventrite (Fig. 13B) large, without glabrous areas, entirely microreticulate superimposed with irregularly shaped setiferous tubercles; the latter appearing reticulately connected in lateral portions; tubercles smaller and shallower at disc; median longitudinal impression deeply impressed, laterally extending into a subtriangular groove; groove without setiferous tubercles. Anepisternum 3 microreticulate with one row of punctures. Hind wings present in all specimens examined, venation not examined.

Legs (Figs 4, 13B, J) approximately as long as body, or very little shorter; coxae large, only procoxae visible in dorsal view; pro- and mesocoxae (Fig. 13B) subglobular (drop-shaped); metacoxae (Fig. 13B) rather obtuse and shallowly elevated, obliquely conoidal; trochanter (Fig. 13B) short, broadly lanceolate, not visible in dorsal view, distal end distinctly pointed; femora and tibiae appearing longitudinally striated by dense cover with very elongate, micro-setiferous tubercles; tibiae distally with rather short and inconspicuous setae; tarsomeres with small scattered setae (Fig. 13J), most

conspicuous at ventral side and near claw insertation; claws (Fig. 13J) large, rather slender, strongly bent; base of each with three teeth, distal one very large (mutilated in specimen figured in 13J).

Ventrite 1 (Fig. 13B) arcuately projected anteriad between hind coxae; microreticulate and tuberceliferous as in metaventrite especially near anterior margin. Ventrites 2 –4 (Fig. 13B) with evenly distributed, subcordiform, setiferous tubercles; interstices almost glabrous; ventrite 5 (Figs 13C, D) evenly covered with short adpressed setae emerging from subcordiform tubercles; lateral projection shallow.

Sternite IX (Figs 13G, H) c. 340  $\mu$ m long, with moderately long anterior strut (distal end broken off in specimen figured in 13G), apical corners rounded, each with one lateroapical seta and one inconspicuous sublateroapical seta; apical margin slightly broadly emarginate; longer paraproct almost reaching apical margin.

Aedeagus (Figs 13K–N) similar to that of *Ancyronyx minutulus* (see Freitag and Jäch 2007: figs 15a, b), but distinctly larger (c. 350 µm long), relatively stouter and without long setae. Median lobe moderately long and moderately slender, with few indistinct pores, subapically straight, not widened, c. 90 µm wide, apically distinctly curved ventrad (lateral view, Figs 13M, N); tip rounded; ventral sac weakly sclerotised except for lateral rim (Figs 13K, M); fibula weakly sclerotised, inconspicuous in transillumination; corona inconspicuous. Phallobase asymmetrical, bent lateroventrad, distinctly longer ventrally, with conspicuous, strongly sclerotised margins; basolateral (penile) apophyses inconspicuous; ejaculatory duct well scleotised and conspicuous in transillumination. Parameres elongately subtriangular, rather short, reaching about basal 0.67 of aedeagus, almost contiguous ventrally, subapically slightly widened ventrad; apices with one apical and one subapical very short setae (Figs 13K, M); basal margin oblique and not conspicuously emarginate (lateral view, Figs 13M, N).

Ovipositor (Fig. 13O, P) c. 410  $\mu$ m long. Stylus slender, rather staight, with various apical sensilla. Coxite moderately stout, distinctly shorter than in specimens of the *Ancyronyx patrolus* species group, but longer than in those of the *A. variegates* group; outer margin concave; all over with several rather short and broad, peg-like spines, increasing in size and density apically at lateral margins; inner margin pubescent; basal portion short. Valvifer moderately longer than coxite; fibula slightly curved.

Secondary sexual characters: Sternite VIII in male short, weakly sclerotised and with very short median strut; in female (Fig. 13I) distinctly longer, more sclerotised than in male medially emarginate; apical corners rounded and with small seate; median portion with dense micro-pubescence. Tergite VIII in female (Fig. 13E) subtriangular, almost as long as wide (c. 210  $\mu$ m long, 230  $\mu$ m wide), with few moderately short setae; condyles large and conspicuous. Tergite VIII in male (Fig. 13F) subsemicircular, distinctly wider than long (c. 170  $\mu$ m long, 230  $\mu$ m wide), shorter than in female, reticulate; apical half with moderately short setae. Ventrite 5 in female (Fig. 13C) subtriangular (c. 230  $\mu$ m long, 400  $\mu$ m wide); in male (Fig. 13D) broadly oval and distinctly shorter (c. 180  $\mu$ m long, 370  $\mu$ m wide).

Adult differential diagnosis. In its colour patterns, *Ancyronyx buhid* resembles *A. patrolus, A. punkti* and especially *A. pseudopatrolus* from Palawan. The new species can

be easily distinguished by the combination of body morphometric and genital characters (body, especially abdomen and elytra, relatively wider (CL/EW c. 2.0; EL/EW c. 1.45) than in other species; legs not distinctly longer than body; coxite of ovipositor moderately stout; aedeagus with straight main piece (not widened subapically), almost contiguous parameres ventrally, very short and few parameral setae.

Larval diagnosis (based on sixth instar). Colour (Fig. 7) dorsally dominantly dark brown except for yellow lateral head, clypeus and labrum, most anterior portion of pronotum and the almost entire first abdominal segment; most specimens additionally with yellowish to pale brown (preterminal) abdominal segment VIII (at least posterior portion) and apex of abdominal segment IX (up to c. posterior 0.15). Legs, mouthparts, ventral head, thorax, and abdomen yellowish to pale brown, but some specimens with darker brown thoracic venter and ventral abdominal segment IX.

HW 0.31 mm; entire larva about 3.1 mm long. Body elongate, wider than that of *A. minerva* and *A. tamaraw*, but similar in the external characters, except for the following: posterolateral projections (Figs 7, 14A) of abdominal segments II–VI usually reaching or slightly overreaching posterior segment margins. Lateral rim of thorax and abdomen with scattered long, trichoid setae. Dorsal sagittal line slightly impressed from prothorax to abdominal segment V and without tubercles.

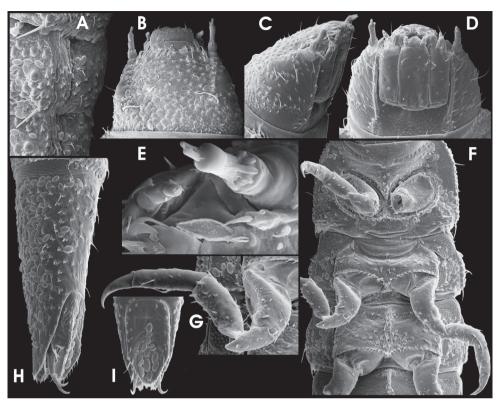
Head (Figs 7, 14B–E) widest posterior 0.3, not subparallel in posterior half, dorsolaterally with a pair of moderately long setae and one pair near the frontoclypeal suture (Fig. 14B); lateral setae of various size, very short to moderately long. Frontal suture inconspicuous; subbasal fringe of clypeus with rather short fasciculate setae. Ventral side (Fig. 14D) dominantly rugulose; basolateral areas and genae (inbetween setae) glabrous (Fig. 14C).

Antenna as in Figs 14D, E, c. ¼ as long as head; scape setae fasciculate; pedicel long; flagellum and sensorium subequal in length.

Labrum (Fig. 14B) with subapical fringe of ramose setae and few lateral trichoid setae. Maxilla (Fig. 14D) with parallel-sided stipes; maxillary palpus (Figs 14D, E) slightly slenderer than in *A. tamaraw*. Labial mentum (Fig. 14D) with lateral margin slightly sinuously curved (concave in posterior half), narrowest at basal 0.2; pair of trichoid setae moderately short (reaching anterior margin), inserted sublaterally at anterior 0.2; pair of subapical lateral setae fasciculate; subbasal pair of setae ramous. Submentum short, not clearly partitioned from somewhat protruding semicircular ligula which is conspicuously covered with setiform microstructures (Fig. 14D); labial palpi as in *A. tamaraw*.

Pro-, meso-, metathorax and legs (Figs 7, 14F) almost as in *A. minerva*. Pronotum with rather inconspicuous small round signa (glabrous areas) in posterior half. Ventral sclerites of thorax (Fig. 14E) rugulose, not glabrous; venter of metathorax with conspicuous sagittal tuberculate ridge (similar to that of the venter in abdominal segment I).

Abdomen (Figs 7, 14A, H, I) without conspicuous dorsosagittal carina except for the anterior half of segment IX (Fig. 12G); squamose setae at posterior rim of segments I–VIII large (Figs 14A, H). Ventral sclerite of segment I with distinct sagittal ridge in anterior half (Fig. 14F), reaching c. 1/2 to 2/3 of segment length. Apex of segment



**Figure 14.** *Ancyronyx buhid* Freitag, sp. n., larva (SEM photographs; not to scale): **A** detail of abdominal segment VII, lateral, with posterolateral projections and spiracles **B** head, dorsal **C** head, lateral **D** head, ventral **E** antenna, frontal **F** thoracic and first abdominal segments, ventral **G** midleg, ventral **H** abdominal segment IX, lateral **I** operculum, ventral.

IX emarginate (sometimes inconspicuous due to apical setae). Operculum (Fig. 14I) almost twice as long as wide, basal portion glabrous.

**Variation between larval instars.** The available prefinal instar specimens vary only slightly from the description above, namely by the relatively slenderer thoracic and abdominal segments, the smaller and rather inconspicuous spiracles near the postero-lateral projection, the slightly broader legs with fewer setae, and the relatively longer lateral setae on thorax and abdomen.

Larval differential diagnosis. The species can most easily be distinguished from any other known *Ancyronyx* larva by the obviously pale first abdominal segment. The general shape and the proportions of the larva of this species resemble those of the *A. patrolus* group, from which it can be additionally distinguished by the anterior yellow band, that is medially narrower (not extended as in several species of the *A. patrolus* group) and the character combination of long sagittal crest of the first abdominal segment venter, slightly impressed dorsosagittal line without protruding tubercles. From the species of the *A. variegatus* group, this larva can be distinguished easily by its spindle-shape habitus (subsemicircular in cross section) and the rather short posterolateral appendages.

**Distribution.** Known only from Mindoro Island where this new species was recorded from various streams in the province of Oriental Mindoro.

**Ecology.** Both, adult and larvae of *Ancyronyx buhid* occur in medium sized, unpolluted rivers in mountainous areas. This suggests an affinity to undisturbed habitats. The relatively highest abundances were found on submerged wood and rough rock surfaces in runs and riffles. Some root packs and partly submerged grass bunches in riffles were also found to be densely colonised with the species. Much more rarely it was found among bottom gravels in runs and calm pools, where specimens were possibly just shifted by drift.

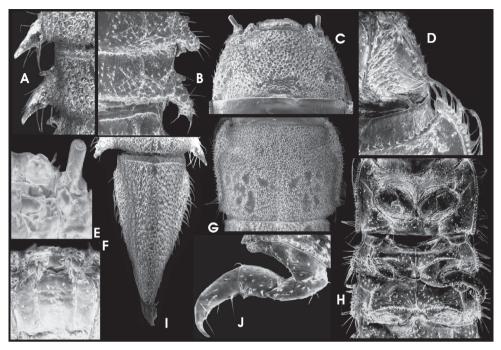
**Remarks.** One male specimen from site "TR2f" varies in regard to the primary and secondary sexual characters, namely the length of tergite VIII, ventrite 5 and aedeagus. Since all other characters do not differ from the type material, this is regarded as an abnormality caused during pupation.

### Ancyronyx schillhammeri Jäch, 1994

http://species-id.net/wiki/Ancyronyx\_schillhammeri Figs 8, 9, 15A–H

*Ancyronyx schillhammeri* Jäch, 1994: 617–619 (adult description), Freitag and Pangantihon 2010: 133–137 (faunistic records).

Material examined. 1  $\bigcirc$  [FR029], 1 L (0.61 [FR030]) (ZSM) "PHIL.: Mindoro, San Teodoro, Tukuran Riv.; small lowld.riv.;riffle &run; woodlitter, gravel; sec.veget.; c.30m asl., 13°25'34"N 120°58'37"E 23.4.1994, leg. Freitag (303a)M"; 3 ♂♂, 1 ♀ (PNM) "PHIL.: Mindoro Oriental, Bongabong, Brgy. Formon, Pastuhan, Tangisan Falls; deep mountain valley, sec. forest, submerged wood, riffle, c.200 m asl., c.12°43'N 121°23'E; 27.10.2011 leg. Freitag (318a)M"; 3 3 3 (CFM) "PHIL:Or.Mindoro, Roxas, Bagumbayan, polluted Magugo River; sec.veget.; submerges wood, run; 6m asl., c.12°35'27"N 121°31'00"E; 05.6.2000 leg. Freitag & Pangantihon (329c)M"; 1 3, 4 ♀♀ (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Baroc River; subm. wood; gravel flood plains; c.12°37'07"N 121°24'06"E, 90m asl; leg. Freitag 1 Apr.2013 (BRf) M";  $4 \frac{3}{6}$ , 7 99 (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente proper, Taugad River; subm. wood; sec.veget.; c.12°37'06"N 121°23'49"E, 100m asl; leg. Freitag 2 Apr.2013(TR1f)M"; 5 ♂♂, 1 ♀ (SMTD, ZSM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Tauga River; subm. wood, run; sec.veget.; c.12°37'18"N 121°22'58"E, c.140m asl; leg. Freitag 28.Nov.2011(TR2f)M"; 1 ♂, 3 ♀♀ (ZMUC) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Tauga Diit, Baroc River tributary Tauga Daka; subm. wood in run; sec.veget.; c. 12°38'05"N 121°19'33"E, c.530m asl; leg. Pangantihon, 23 Jan. 2013 (TDR3f)M"; 1 ♀ (CFM) "PHIL:Or.Mindoro, Roxas, Brgy. San Vicente, Sitio Quirao, Baroc River tributary Hinundugan River; subm. wood; c.



**Figure 15.** *Ancyronyx schillhammeri* Jäch, 1994, larva (SEM photographs) not to scale): **A** detail of abdominal segments II–III, dorsal, with posterolateral projections and spiracles **B** detail of abdominal segments VI–VII, ventral, with posterolateral projections and spiracles **C** head, dorsal **D** detail of head and prothorax, ventral **E** antenna, ventral **F** maxillae and labium, ventral **G** pronotum with signa, dorsal **H** thorax, ventral **I** abdominal segment IX, dorsal **J** midleg, ventral.

12°36'23"N 121°23'29"E, c. 118m asl; leg. Pangantihon, 22 Jan.2013 (HR1f)M"; 1  $\bigcirc$  (CFM) "PHIL:Oc.Mindoro, San Jose, Brgy. "Central" Purok Tunnel, Busuanga River; rural,cogon gras roots, riffle & run, c.73 m asl., c.12°27'51"N 121°02'08"E; 07.VI.2012 leg. Freitag & Pangantihon (330b)M".

Larval diagnosis (based on a single presumably sixth instar specimen). Colour in last instar larva predominantly brown as in Fig. 9; dorsal head darkest to almost black at pronotal disc; lateral head, antennae, anterior and lateral pronotal margins, legs (except for tip of claw), lateral abdominal segment margins and conical projections, posterior abdominal tip and areas around the sagittal line (especially thoracic area) distinctly paler, yellowish to pale brown. Ventral side entirely pale except for pale brown gula, maxillae and labium; ventral part of genae conspicuously dark brown.

HW c. 0.60 mm; entirely c. 3.8 mm long.

Body shape of the *A. variegatus* group type, generally very similar to that of *A. procer-us* (comp. Freitag and Balke 2011: 72–75) in the external habitus, except for the following: Posterolateral projections (Figs 9, 15A, B) of abdominal segments IV– VIII slightly broader and stouter; spiracles distinctly larger, very prominent; entire lateral margin with distinct long trichoid setae; tubercles much more prominent (especially at dorsal side).

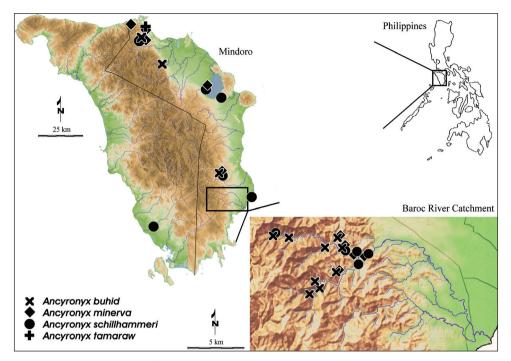


Figure 16. Collecting sites of the *Ancyronyx* species in Mindoro Island, including enlarged map of the Baroc River Catchment.

Head (Figs 15C-F) distinctly widest basally, slightly conical anteriad; without median pointed projection at frons; the pair of sublateral anterior projections between antenna and clypeus rather shallow (Fig. 15C). Frons moderately densely and equally covered with prominent setiferous tubercles; setae very short. Genae rugose, with irregular depressions, ridges, and scattered tubercles (Fig. 15D); lateral glabrous area with stemmata elongately subtriangular (Fig. 15D). Antenna (Fig. 15E) less than half as long as head. Scape short, slightly longer than wide, with subapical fringe of stout sensilla; pedicel cylindrical less than three times as long as scape and c. three times as long as wide, with few apical sensilla; flagellum and sensorium as in A. procerus. Ventral side (Fig. 15F) with well-developed longitudinal crests bordering the stipes. Labrum broad, c. 3.5 times as wide as long; lateroapical edges rounded; entire visible dorsal surface with small setiferous tubercles. Maxilla (Fig. 15E, F) almost as in A. procerus. Labium (Figs 15E, F) with mentum widest in apical half; pair of moderately long trichoid setae inserted sublaterally at anterior 0.3; some additional trichoid setae present at lateral margin in apical half; pair of apicolateral teeth inserted at a distinct subapical crenation; submentum straight, without conspicuous median ridge, apically broadly concave.

Prothorax (Fig. 15G, H) slightly wider than long; tergum with irregularly shaped and round signa in posterior half; median and submedian pairs clearly defined by bordering tubercles, not fused (Fig. 15G). Venter of prothorax (Fig. 15H) similar to that in *A. procerus*, but anterior sclerites distinctly shorter, oblique, not subtriangular; anterior margin with conspicuous setiferous tubercles; anterior and lateral sclerites partly fused near anterior prothorax margin; transverse sutures dividing the lateral sclerites distinctly ending before lateral margin. Venter of meso- and metathorax (Fig. 15H) with more conspicuous setiferous tubercles particularly at posterior margins of anterior sclerites.

Legs (Figs 15H, J) proportioned as in *A. procerus*, but tubercles and setae larger and more distinct. Subbasal tooth of claws long and trichoid, overreaching tip of claw.

Abdominal terga (Figs 9, 15A, B, I) with slightly depressed groove along sagittal line at least from 1<sup>st</sup> up to 4<sup>th</sup> segment; posterior terga margins with rim of squamose setae. Posterior venter margins with rim of trichoid setae. Segment IX (Fig. 15I) dorsally with shallow sagittal crest formed by densely arranged tubercles bearing large trichoid setae; apex widely rounded to slightly truncate; ventral side rugose, not glabrous. Operculum without longitudinal ridges, entirely rugose and covered with conspicuous scattered setae.

Larval differential diagnosis. The larvae of *Ancyronyx schillhammeri* are easily distinguishable from all other known *Ancyronyx* larvae of Mindoro by their larger size, the somewhat dorsoventrally depressed habitus, the much larger and protruding posterolateral appendages, as typical for the *A. variegates* species group. Among this species group, it resembles the larva of *A. procerus* in colour, but can be clearly distinguished by the absence of the pointed projections at median frons, the more shallow projections between antenna and clypeus, the larger and more protruding spiracles, the conical head shape, and the surfaces of head, thorax, and abdomen that are densely covered with larger tubercles bearing long conspicuous setae. From *A. helgeschneideri* it is easily distinguishable by the darker colour, the pale dorsosagittal stripe, the dark dorsal abdominal segment IX and the broader and conical head.

**Distribution.** Only known from Oriental Mindoro and one locality of Occidental Mindoro near San Jose.

**Ecology.** *Ancyronyx schillhammeri* occurs exclusively on submerged wood. Decaying pale light woods appear to be preferred by the species. At the sites of the Baroc River catchment, which were sampled regularly throughout the year, the abundance of this species was found to increase distinctly towards the end of the dry season (February to April) and declines rapidly with the beginning rainy season, presumably due to wash out. It is found in both habitat types: clean, cool and torrent rhithral creeks and rivers as well as warm, mesosaprobic lowland streams. This suggests less specific ecological requirements in terms of stream hydraulics, water temperature, and water quality.

### Key to the adult Ancyronyx species of Mindoro

1	Elytra with yellowish "X"-shaped colour pattern (Fig. 8); combined length
	of pronotum and elytra (CL) $\geq$ 1.8 mm; legs very long ( $\geq$ 1.2 times of body
	length)
_	Elytra with four isolated yellowish colour patches; $CL \le 1.4$ mm; legs long to
	moderately long (≤ 1.1 times of body length) <b>2</b>

2	Body about half as wide as long (CL/EW c. 2.0), elytral shoulder distinctly
	wider than pronotum; posterior yellowish elytral patches more or less round,
	not distinctly elongate
-	Body less than half as wide as long (CL/EW $\ge$ 2.2), elytral shoulder about
	as wide as pronotum; posterior yellowish elytral patches elongate, distinctly
	longer than wide
3	Pronotum anteriorly with transverse white to yellowish band; femora entirely
	dark coloured; anterior yellowish elytral patches large, extending over more
	than 3/4 <sup>th</sup> of elytral width (Fig. 2)
_	Pronotum entirely dark, without transverse band; femora predominantly yel-
	lowish coloured; anterior yellowish elytral patches small, extending over 1/2 or
	less of elytral width (Fig. 3)

# Key to the larvae of Ancyronyx species of Mindoro

1	Body flattened dorsoventrally (depressed), only slightly vaulted. Posterolat- eral abdominal projections large, conical, with tip posterolaterad directed. Dorsal colour predominantly dark brown, with distinct pale sagittal stripe in
	anterior half (Fig. 9)
	Body spindle-shaped, elongate, subsemicircular in cross section. Posterolat-
-	
	eral abdominal projections small, lobate, with tip posteriad directed. Pre-
	dominant dorsal colour brown, with transverse pale yellowish bands or dots,
	without any pale longitudinal stripe (Figs 5–7)2
2	First abdominal segment almost entirely yellowish, appearing as an obvious
	pale band. Dark portion of the pronotum medially extended anteriad, yel-
	lowish anterior pronotal band medially narrower. (Fig. 7)A. buhid
_	First abdominal segment almost entirely brownish dark, no obvious pale band
	present at the dorsal abdomen. Anterior pronotal yellowish band regularly
	shaped or extending medially, not narrower along the midline (Figs 5–6)
	( <i>A. minerva</i> species subgroup) <b>3</b>
3	Anteriomedian portion of the dorsal head yellowish pale, surrounded by dark
5	areas. Last abdominal segment c. 2.3 times as long as wide, with pale apical and
	median areas and distinctly dark portion inbetween (Fig. 6) <i>A. tamaraw</i>
-	Entire dorsal head disc brownish dark. Last abdominal segment c. 2.2 times as
	long as wide, any pale dorsal colour pattern lacking or limited to tip (Fig. 5)
	A. minerva

# Updated check list of the Philippine species of Ancyronyx

- 1. Ancyronyx buhid Freitag, 2013 (Mindoro)
- 2. Ancyronyx helgeschneideri Freitag & Jäch, 2007 (Palawan, Busunga)

- 3. Ancyronyx minerva Freitag & Jäch, 2007 (Palawan, Mindoro)
- 4. Ancyronyx minutulus Freitag & Jäch, 2007 (Palawan)
- 5. Ancyronyx montanus Freitag & Balke, 2011 (Palawan)
- 6. Ancyronyx patrolus Freitag & Jäch, 2007 (Palawan, Busuanga)
- 7. Ancyronyx procerus Jäch, 1994 (Busuanga, Borneo, Vietnam)
- 8. Ancyronyx pseudopatrolus Freitag & Jäch, 2007 (Palawan)
- 9. Ancyronyx punkti Freitag & Jäch, 2007 (Palawan)
- 10. Ancyronyx schillhammeri Jäch, 1994 (Mindoro)
- 11. Ancyronyx sophiemarie Jäch, 2004 (Sibuyan)
- 12. Ancyronyx tamaraw Freitag, 2013 (Mindoro)

# Discussion

During the last two decades, the Philippine Islands have received increasing attention in biodiversity research, not least because they are classified as a major biodiversity hotspot in global scale (Myers et al. 2000). However, for several taxa including Elmidae and other freshwater macroinvertebrates, it still requires substantial efforts to record and to describe the majority of species and their distribution.

Four species of *Ancyronyx* are now recognised and formally described from Mindoro Island based on the study of a copious collection of museum specimens and the material retrieved from a comprehensive survey of the Baroc River Catchment in southern Mindoro. Only one of them, *A. minerva*, is recorded beyond Mindoro. *A. schillhammeri* and *A. buhid* appear to be endemic to the island. The high rate of island endemism reflects the biogeographic history of the island. Mindoro is a remnant of a fragment of the Eurasian continental margin and is not part of the Luzon arc of islands of marine volcanic origin (Hall 1998). Despite its recent close vicinity to Luzon, the two islands remained largely isolated in the Quaternary, even during Pleistocene when low sea levels have formed land bridge interconnections of several Philippine islands, but presumably Greater Palawan, Mindoro, and Greater Luzon remained separated based on Pleistocene sea-level low stands represented by the 120 m isobath (Sathiamurthy and Voris 2006).

Therefore, it requires more in depth phylogenetic and biogeographic studies to explane the distribution of *A. minerva* at both sides of the Mindoro Strait.

The phylogenetic relationship of *A. buhid* with other members of the genus is still ambiguous. Several taxa (probably new species) from the Philippines and Sulawesi that resemble *A. buhid* still await their description (unpublished material of the author and at NMW). After this material has undergone detailed study and molecular genetic analysis sound conclusions might be drawn.

The fact that *Ancyronyx* (and very most other Elmidae) live permanently under water and respire by a microplastron (Kodada and Jäch 2005) makes them sensitive to water pollution. The vivid and specific colour patterns of adult *Ancyronyx* species enabling an easy identification, as well as the availability of regional identification keys for both, larvae and adults, allow their potential use as bioindicators. Among the

species of Mindoro, *A. schillhammeri* was recorded from clean to moderately polluted streams making it unsuitable as a bioindicator. The remaining Mindoro species seem to be ecologically adapted to clean and rather undisturbed waters. However, *Ancyronyx tamaraw* is too rare to serve as good bioindicators and *A. minerva* is occasionally detected in slightly polluted streams in low abundances (Freitag and Pangantihon 2010), suggesting a low indicator strength. Therefore, *A. buhid* in particular has the highest potential value to be used as saprobic indicator. Its frequent occurrence in suitable habitats and the easy identification by the distinguishing elytral colour pattern and broad elytral shoulders in adults, as well as the unique yellow abdominal pattern in larvae, make it a suitable tool for biomontoring, even for non-entomologists. However, ecological evaluations of larger scale are needed to confirm these preliminary findings.

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# References

- Agencourt Bioscience (2006) Agencourt CleanSEQ Dye-Termintor Removal, protocol 000600v32. Agencourt Bioscience Corporation, Beverly, 12 pp.
- Ambal RGR, Duya MV, Cruz MA, Coroza OG, Vergara SG, de Silva N, Molinyawe N, Tabaranza B (2012) Key Biodiversity Areas in the Philippines: Priorities for Conservation. Journal of Threatened Taxa 4 (8): 2788–2796. doi: 10.11609/JoTT.o2995.2788-96
- Baraclough TG, Hogan JE, Vogler AP (1999) Testing whether ecological factors promote cladogenesis in a group of tiger beetles (Coleoptera: Cicindelidae). Proceedings of the Royal Society of London B 266: 1061–1067. doi: 10.1098/rspb.1999.0744
- Brown HP (1972) Aquatic dryopoid beetles (Coleoptera) of the United States. Biota of freshwater ecosystems identification manual no. 6. Water Pollution Control Research Series, EPA, Washington D.C., 82 pp.
- Curtis J (1830) British Entomology; being illustrations and descriptions of the genera of insects found in Great Britain and Ireland: Containing coloured figures from nature of the most rare and beautiful species, and in many instances of the plants upon which they are found. London, vol. 7: 290–337.
- Erichson WF (1847) Naturgeschichte der Insecten Deutschlands. Nicolaische Buchhandlung, Berlin, 968 pp.
- Freitag H (2012) Ancyronyx jaechi sp.n. from Sri Lanka, the first record of the genus Ancyronyx Erichson, 1847 (Insecta: Coleoptera: Elmidae) from the Indian Subcontinent, and a world checklist of species. Zootaxa 3382: 59–65. http://www.mapress.com/zootaxa/2012/f/z03382p065f.pdf
- Freitag H, Balke M (2011) Larvae and a new species of Ancyronyx Erichson, 1847 (Insecta, Coleoptera, Elmidae) from Palawan, Philippines, using DNA sequences for the assignment of the developmental stages. ZooKeys 136: 47–82. doi: 10.3897/zookeys.136.1914
- Freitag H, Jäch MA (2007) The genus Ancyronyx Erichson, 1847 (Coleoptera, Elmidae) in Palawan and Busuanga, (Philippines) with description of six new species. Zootaxa 1590: 37– 59. http://www.mapress.com/zootaxa/2007f/z01590p059f.pdf
- Freitag H, Pangantihon C (2010) Aquatic Coleoptera and Heteroptera of the Lake Naujan National Park, Mindoro Oriental, the Philippines. Philippine Scientist 47: 126–173. http:// www.philjol.info/philjol/index.php/PSCI/article/view/2424
- Hadley A (2008) http://hadleyweb.pwp.blueyonder.co.uk/CZM/combinezm.htm Version of 18<sup>th</sup> April 2008.
- Hall R (1998) The plate tectonics of Cenozoic SE Asia and the distribution of land and sea. In: Hall R, Holloway JD (Eds) Biogeography and Geological Evolution of SE Asia. Backhuys Publishers, Leiden, 99–131.
- Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41: 95–98.
- Hilsenhoff WL (1982) Using a biotic index to evaluate water quality in streams. Wisconsin Department of Natural Resources Technical Bulletin 132: 22 pp.
- Hinton HE (1941) New genera and species of Elmidae. Transactions of the Royal Entomological Society of London 91(B) (3): 65–104.

- Jäch MA (1982) Neue Dryopoidea und Hydraenidae aus Ceylon, Nepal, Neu Guinea und der Türkei. (Col.). Koleopterologische Rundschau 56: 89–114.
- Jäch MA (1994) A taxonomic review of the Oriental species of the genus *Ancyronyx* Erichson, 1847 (Coleoptera, Elmidae). Revue suisse de Zoologie 101 (3): 601–622. http://biostor. org/reference/115102
- Jäch MA (2004) Descriptions of two new species of *Ancyronyx* Erichson (Insecta: Coleoptera: Elmidae). Annalen des Naturhistorischen Museums in Wien 105 B (2003): 389–395.
- Kodada J, Jäch MA (2005) 18.2. Elmidae Curtis, 1830. In: Beutel RG, Leschen RAB (Eds) Handbook of Zoology, Volume IV (Part 38), Coleoptera, Beetles, Volume 1: Morphology and Systematics (Archostemata, Adephaga, Myxophaga, Polyphaga partim). Walter de Gruyter, Berlin, New York, 471–496.
- Moog O, Jäch MA (2003) Elmidae. In: Moog O (Ed) Fauna Aquatica Austriaca, edition 2002. Wasserwirtschaftskataster, Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft, Wien, 68 pp.
- Myers N, Mittermeier RA, Mittermeier CG, da Fonseca GAB, Kent J (2000) Biodiversity hotspots for conservation priorities. Nature 403: 853–858. doi: 10.1038/35002501
- Ong PS, Afuang LE, Rosell-Ambal RG (Eds) (2002) Philippine Biodiversity Conservation Priorities: A Second Iteration of the National Biodiversity Strategy and Action Plan. Department of Environment and Natural Resources –Protected Areas and Wildlife Bureau, Conservation International Philippines, Biodiversity Conservation Program University of the Philippines Center for Integrative and Development Studies, and Foundation for the Philippine Environment, Quezon City, Philippines, 113pp.
- Ronquist F, Teslenko M, van der Mark P, Ayres D, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61 (3): 539–42. doi: 10.1093/sysbio/sys029
- Sathiamurthy E, Voris HK (2006) Maps of Holocene Sea Level Transgression and Submerged Lakes on the Sunda Shelf. The Natural History Journal of Chulalongkorn University, Supplement 2: 1–43.
- Simon C, Frati F, Beckenbach A, Crespi B, Liu H, Flook P (1994) Evolution, weighting and phylogenetic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers. Annals of the Entomological Society of America 87: 651–702.
- Tavaré S (1986) Some probabilistic and statisical problems on the analysis of DNA sequences. Lectures in Mathematics in the Life Sciences 17: 57–86.
- Thompson JD, Higgins DG, Gibson TJ (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. Nucleic Acids Research 22: 4673–4680. doi: 10.1093/nar/22.22.4673
- Qiagen (2002) DNeasy Tissue Kit Handbook 05/2002. Hilden, Germany, 43 pp.

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RESEARCH ARTICLE



# A review of the New World species of the parasitoid wasp *lconella* (Hymenoptera, Braconidae, Microgastrinae)

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### Abstract

The New World species of *Iconella* (Hymenoptera: Braconidae, Microgastrinae) are revised. *Iconella andydeansi* Fernández-Triana, **sp. n.**, *I. canadensis* Fernández-Triana, **sp. n.**, and *I. jayjayrodriguezae* Fernández-Triana, **sp. n.**, are described as new. *Iconella isolata* (Muesebeck, 1955), **stat. r.**, previously considered as a subspecies of *I. etiellae* (Viereck, 1911), is here elevated to species rank. All species have different, well defined geographic distributions and hosts. Taxonomic keys are presented in two formats: traditional dichotomous hardcopy versions and links to electronic interactive versions (software Lucid 3.5). Numerous illustrations, computer-generated descriptions, distributional information, host records (mostly Lepidoptera: Crambidae

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and Pyralidae), and DNA barcodes (where available) are presented for every species. Phylogenetic analyses of the barcoding region of COI indicate the possibility that *Iconella* is not monophyletic and that the New World species may not form a monophyletic group; more data is needed to resolve this issue.

#### **Keywords**

*Iconella*, Microgastrinae, New World, taxonomic review, host caterpillars, DNA barcoding, Area de Conservacion Guanacaste, parasitoid wasps

### Introduction

The genus *Iconella* was erected by Mason (1981) to accommodate a group of *Apanteles* species with a sinuated vein cu-a in the hind wing, a character he interpreted as plesiomorphic among Microgastrinae. *Iconella* includes all species of Nixon's *sundanus*-group and part of the *merula*-group (Nixon 1965, Whitfield 1997). It is cosmopolitan, with 33 described species, most of them found in the Palearctic (Yu et al. 2012). All species with known hosts are solitary parasitoids of microlepidoptera, especially concealed feeders (Whitfield 1997); one species, *Iconella isolata* (Muesebeck, 1955) has been extensively studied in the tropics for biological control of a pest caterpillar (e.g. Bennett 1960, Bartlett et al. 1978).

Until now, only one species was known from the New World, *Iconella etiella* (Viereck, 1911). However, ongoing research on the Microgastrinae fauna of Area de Conservacion de Guanacaste (ACG) in northwestern Costa Rica (Smith et al. 2008, Janzen et al. 2009) and study of specimens housed in the Canadian National Collection of Insects (CNC) in Ottawa, Canada, have both revealed several new species for the Western Hemisphere. They are described here, and a key to all species in the New World is provided.

It should be noted that Achterberg (2002) did not consider *Iconella* as a valid genus and transferred its species (as well as those of other genera) back to *Apanteles*. However, that decision has not been universally accepted (e.g. Fernández-Triana 2010, Broad et al. 2012) and is still a subject of debate. In this paper we treat *Iconella* as a valid genus on its own.

### Methods

*Iconella* is rare in collections, and even when present, specimens tend to be categorized as "unidentified Microgastrinae". In the New World, with the exception of one species, the genus seems to be rare in nature as well. Even long-term and comprehensive rearing programs –such as those in ACG, Costa Rica- have recovered relatively few specimens. This study is mostly based on the examination of unidentified *Iconella* specimens housed in the CNC, representing close to 40 specimens; 11 specimens reared by the ACG inventory -and housed in the CNC and the Illinois Natural History Survey, Champaign, Illinois, United States (INHS); one specimen from the Laurentian Forestry Center, Ste.-Foy, Quebec, Canada (LFS); and the holotype of *I. etiellae* from the Smithsonian Institution, Washington DC, United States (NMNH).

Morphological terms and measurements of structures are mostly as used by Mason (1981), Huber and Sharkey (1993) and Whitfield (1997). Non-morphological characters are also provided in the key whenever available (e.g., host species, geographical distribution). Those features are included in brackets at the end of the corresponding couplet and are intended as supplementary information that can help with identification.

The species descriptions are based mostly on the holotype female, with other specimens studied (when available) for intraspecific variation. When the holotype could not be examined (*I. isolata*) or it was a male (*I. etiellae*), female specimens were used for these redescriptions.

Lucid 3.5.4 (http://www.lucidcentral.com/) software was used to generate automatic descriptions of the species and to prepare Lucid identification keys. A dataset of 20 characters and 126 character-states was used to provide uniform descriptions for all species treated. Description format includes one sentence per character, with the character (in italics) mentioned first and the character-state following after a colon, e.g., "*Pterostigma color:* mostly brown, with yellowish-white spot at anterior  $0.2 \times$ ".

A map with the distribution of all New World species of *Iconella* was generated using SimpleMappr (Shorthouse 2010).

In the taxonomic treatment of species, "Specimens Examined" presents the specimen's information in the following format: "Number of females/males, acronym of the storing institution between parentheses, COUNTRY: State/Province, city, other locality details, coordinates (in Decimal Degrees, abbreviated as "Lat:" and "Long:"), date, collector name, biological information on host (starting with "ex"), ACG database codes (in the format "yy-SRNP-xxxxx" or "DHJPARxxxxxx", with the former being the voucher code of the host and the latter being the voucher code of the parasite). For states of the United States and for Canadian provinces/territories, acronyms consisting of two capital letters are used, following Canada Post (http://www.canadapost.ca/tools/pg/manual/PGaddress-e.asp).

DNA barcodes for all specimens that were barcoded were obtained using DNA extracts prepared from single legs using a glass fibre protocol (Ivanova et al. 2006). Extracts were re-suspended in 40  $\mu$ l of dH2O, and a 658-bp region near the 5' terminus of the COI gene was amplified using standard primers (LepF1–LepR1) following established protocols (Smith et al. 2006, 2007, 2008). If the initial 658 bp amplification was not successful, composite sequences were generated using internal primers (primers are as detailed in Smith et al. (2008)). All available DNA barcodes for *Iconella* specimens from the New World and the Palearctic are available on the Barcode of Life Data System (BOLD, www.boldsystems.org/), along with sequences from *Apanteles* and *Dolichogenidea*, which were used as outgroups in the phylogenetic analyses (dx.doi.org/10.5883/ DS-ASICON1). Collection information and accessions (BOLD and GenBank) for all sequences were already published in a previous paper (Smith et al. 2013).

DNA barcode sequences were aligned in Geneious Pro 6.0.5 (Drummond et al. 2011) using the default settings for a MUSCLE alignment. Because of the high AT content characteristic of Insect and Hymenoptera mitochondrial DNA, sequence divergences were calculated using the TN93model (Tamura and Nei 1993) and a neighbor-joining

(NJ, Saitou and Nei 1987) tree of distances was generated using Geneious Pro 6.0.5 (Drummond et al. 2011) to provide a graphical representation of the species divergences.

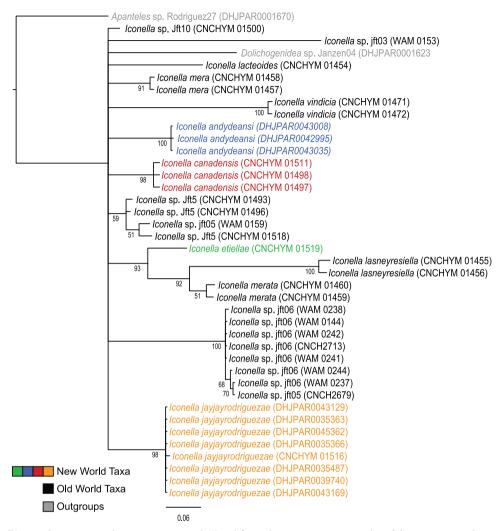
Many of the *Iconella* specimens in BOLD were collected before 1979. Characteristic of such moderately-aged specimens, the COI fragments generated were less (~160bp) than the full length characteristic of the standard DNA barcode (>600bp) (Fernández-Triana et al. 2011). Therefore, more than half of the sequences used in the analyses described below were less than 300bp, and 44% were less than 200bp. Phylogenetic comparisons of fragments this small, and involving cases where there is even lower overlap will be compromised. However, small COI fragments (<200bp) are not devoid of phylogenetic signal and have been used to successfully identify species (Fernández-Triana 2010, Fernández-Triana et al. 2011, Hajibabaei et al. 2006).

To investigate the phylogenetic relationships of the species, the aligned dataset was analyzed in MrBayes v. 3.2.1 (Ronquist and Huelsenbeck 2003). Model selection was based on the Akaike Information Criterion as implemented in JModelTest v.2.1.1 (Darriba et al. 2012). Two independent analyses with 4 chains each were run in parallel for 10 million generations under a GTR+I+G model. The parameter trace files of each run were observed in Tracer v.1.5 (Rambaut and Drummond 2009) to verify that the runs had converged on the same stationary distribution, and to decide on the appropriate number of generations to discard as burn-in. A 50% majority rule consensus tree was constructed from the 18 million post-burn-in generations in Geneious Pro 6.0.5. The above protocol was followed for additional analyses in which 1) all 3<sup>rd</sup> codon positions were removed from the dataset, 2) characters with more than 2% missing data were removed, and 3) sequences that were less than 547 base pairs long were eliminated. A Maximum Likelihood (ML) analysis was also run in RAXML v. 7.3.4 (Stamatakis 2006) under a GTR+I+G model. 1000 bootstrap (BS) replicates were run and the BS support values were then drawn onto the best-scoring ML tree. A parsimony analysis was conducted in TNT v.1.1 (Goloboff et al. 2008) using the "aquickie.run" script provided with the program.

### Results

As with many other taxa, the generic status of *Iconella* will only be solved when a comprehensive phylogeny of Microgastrinae is carried out. In the meantime we think is best to keep it as a valid genus, based on the available morphological evidence. Mason (1981) already mentioned the sinuated vein cu-a in the hind wing as a plesiomorphic character that suggests the unique status of *Iconella* among similar genera. Besides that, we also consider the presence of a median longitudinal carina on the propodeum (or the secondary loss of that carina, which occurs in some Paleartic species but not in the New World species) as a strong support for the generic status of *Iconella* –in contrast with *Apanteles*, which (*sensu* Mason 1981) almost always has carinae defining a complete or partial areola on the propodeum, but never has a median longitudinal carina on the propodeum.

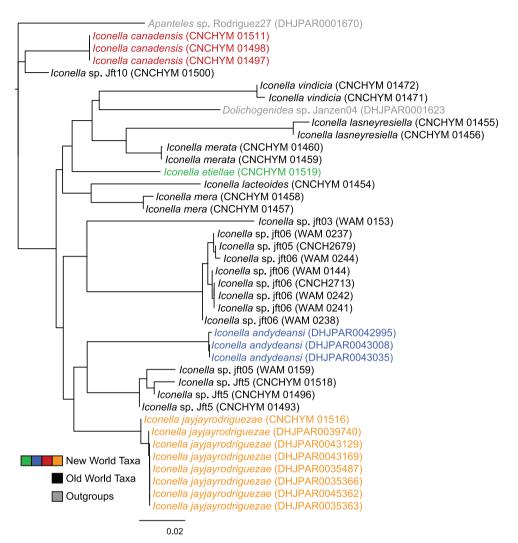
Phylogenetic analyses of the COI DNA, however, are inconclusive as to whether the genus *Iconella* is monophyletic (Figs 1 and 2). All Bayesian (only the tree resulting



**Figure 1.** Majority rule consensus tree calculated from the posterior tree samples of the Bayesian analysis of the full *Iconella* COI fragment dataset. Values below the branches are posterior probabilities. The New World species are individually color coded.

from analysis of the full dataset is shown in Fig. 1), ML (tree not shown), and parsimony (tree not shown) analyses failed to recover monophyly of the genus. However, the model-based methods do not offer strong support against monophyly either. The highest posterior probability (PP) supporting non-monophyly was 0.55 when 3<sup>rd</sup> codon positions were removed from the Bayesian analysis, and BS support against monophyly in the ML analysis was only 0.21.

We recognize five species of *Iconella* in the New World, including the three new species described in the present paper, and one that was previously considered as a subspecies (Muesebeck 1955) but is here elevated to species. We are also aware of



**Figure 2.** Neighbor-joining tree of the full *Iconella* COI fragment dataset using the TN93model. The New World species are individually color coded.

an additional sixth species, represented by two specimens in poor condition from British Columbia (Western Canada), deposited in the CNC. They were mentioned in Fernández-Triana (2010) as "*Iconella* sp. 2" and are different from all other New World species of the genus. However, until more material is found, it is impossible to describe that species.

All of the described species have a different, well delimited geographic range (Fig. 3) and also differ in their known hosts. Most of the host species in the Western Hemisphere belong to the Lepidoptera families Pyralidae and Crambidae (with appropriate caution that what was called Pyralidae in the past is in part called Crambidae at present). The NJ tree (Fig. 2) as well as Bayesian (Fig. 1), ML, and parsimony based

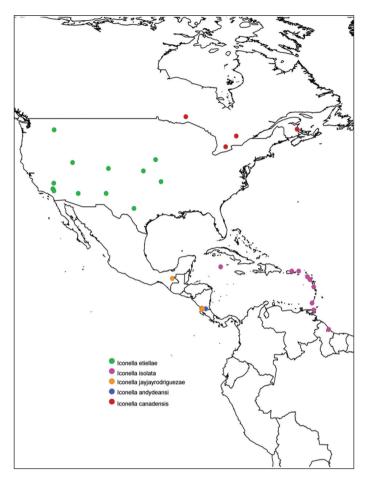


Figure 3. Distribution of New World species of Iconella. Species are individually color coded.

phylogenetic analyses of the barcodes support the validity of all New World species recognized herein (there is no molecular data available for *I. isolata*). However, the barcoding dataset did not contain a strong enough phylogenetic signal to resolve the phylogenetic relationships among *Iconella* species.

While these DNA barcodes and short COI fragments (mini-barcodes (Hajibabaei et al. 2006, Meusnier et al. 2008)) were sufficient to unambiguously separate the species of *Iconella*, more collections and longer DNA sequences will be needed to test the monophyly (or lack thereof) of *Iconella*.

# Key to the New World species of Iconella

1 Propodeum mostly smooth and polished (as in Fig. 21), except for sparse punctures on the anterior  $0.2 \times$  of propodeum, and rather small carinae ra-

diating from median longitudinal carina; metatibia mostly yellow, at most with very small and faint brown spot on posterior 0.1 or less, and metatarsus mostly yellow, except for brown area on posterior half of first tarsomerus (Figs 17, 19, 23, 25); fore wing with most veins transparent or white, vein margins of same color than interior of vein (Figs 18, 24) ......2 Propodeum with anterior  $0.2 \times$  covered with punctures, posterior 0.8 covered with mix of punctures, striated sculpture and carinae (as in Fig. 15); metatibia with brown to black coloration on posterior 0.2-0.4, and metatarsus mostly dark brown, except for yellowish area on anterior half of first tarsomerus (as in Fig. 33); fore wing with at least some veins with thin brown Pterostigma almost completely brown, with only small whitish spot anteriorly (Fig. 18); humeral complex half yellow, half brown; profemur almost completely dark brown (yellow area absent or limited to posterior 0.2); interocellar distance 2.4 × or more posterior ocellus diameter (Fig. 20); mediotergite 2 width at posterior margin 4.6 × or less its length (Fig. 22); larger species, body length (head to apex of metasoma) 3.0 mm or more, and fore wing length 3.3 mm or more. [Western and central United States: AR, AZ, CA, CO, IA, NM, OK, and WA. Hosts: Etiella zinckenella, Olycella junctolineella, Psorosina hammondi, and Ufa rubedinella (Pyralidae)] .....

*Iconella etiellae* (Viereck, 1911) Pterostigma mostly transparent or whitish, with only thin brown margins (Fig. 24); humeral complex yellow to white; profemur mostly yellow, dark brown area limited to anterior 0.2 or less; interocellar distance 2.1 × or less posterior ocellus diameter; mediotergite 2 width at posterior margin 5.0 × or more its length (Fig. 26); smaller size, body length (head to apex of metasoma) 3.0 mm or less, and fore wing length 3.2 mm or less. [Caribbean islands and northern part of South America: British Virgin Islands, Cayman Islands, Dominica, Grenada, Guyana, Montserrat, Puerto Rico, Saint Kitts & Nevis, Trinidad & Tobago. Host: *Ancylostomia stercorea* (Pyralidae)] .....

*Iconella isolata* (Muesebeck, 1955), stat. r. Ocular-ocellar line 1.6 × posterior ocellus diameter; humeral complex half yellow, half brown; mediotergite 1 width at anterior margin 2.2 × or less its width at posterior margin (Fig. 13); ovipositor sheaths length 0.8 × or less metatibial length (Fig. 12); larger species, body length (head to apex of metasoma) 3.5 mm or more (rarely 3.2 mm) and fore wing length 3.5 mm or more; an extra-tropical species distributed in North America north of 40° N (Canada). [Eastern Canada: NB, ON, and QC. Host: *Epinotia solandriana* 

*Iconella canadensis* Fernández-Triana, sp. n. Ocular-ocellar line  $2.0 \times \text{or}$  more posterior ocellus diameter; humeral complex fully yellow to white; mediotergite 1 width at anterior margin  $3.1 \times \text{or}$  more its width at posterior margin (Figs 9, 36); ovipositor sheaths length

(Tortricidae) and, likely, Acrobasis betulella (Pyralidae)].....

2

3-

 $1.1 \times$  metatibial length (Figs 6, 32); smaller size, body length (head to apex of metasoma) 3.0 mm or less, and fore wing length 3.3 mm or less; tropical species from Central America south of 17° N (Mexico and Costa Rica)......4 4 Profemur mostly yellow, dark brown area limited to anterior 0.2 or less; meso- and meta- femora mostly dark brown, with proximal 0.1-0.2 yellow to orange; mesoscutellar disc sculpture centrally smooth with few, scattered punctures near margins (Fig. 36); mediotergite 2 width at posterior margin  $4.1 \times \text{or}$  less its maximum length medially (Fig. 36); body length (head to apex of metasoma) 2.9-3.0 mm; fore wing length 3.2-3.3 mm. [Costa Rica (ACG) and Mexico (Chiapas). Host: undescribed species of Phycitinae (Pyralidae)]..... Profemur dark brown on anterior half, yellow on posterior half; meso- and meta- femora usually fully dark brown to black; mesoscutellar disc sculpture mostly with punctures scattered all over disc surface (Fig. 9); mediotergite 2 width at posterior margin  $4.4 \times$  its maximum length medially (Fig. 9); body length (head to apex of metasoma) 2.7-2.8 mm; fore wing length 3.0 mm. [Costa Rica (ACG). Host: undescribed species of Spilomelinae (Crambidae)]...... Iconella andydeansi Fernández-Triana, sp. n.

#### Taxonomic treatment of species, in alphabetical order

#### Iconella andydeansi Fernández-Triana, sp. n.

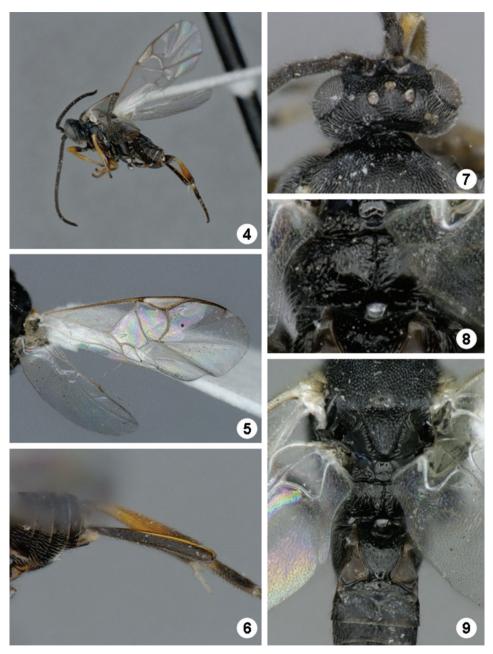
http://zoobank.org/C9173C9E-1E3C-46A8-A459-97DDF2ABF1DF http://species-id.net/wiki/Iconella\_andydeansi Figures 4–9

**Type locality.** COSTA RICA, Alajuela, Area de Conservacion Guanacaste, Sector Rincon Rain Forest, Camino Rio Francia, 410m. Lat: 10.90425, Long: -85.28651.

**Holotype.** ♀, CNC. First label: DHJPAR0043035. Second label: Voucher: D.H.Janzen & W.Hallwachs, DB: http://janzen.sas.upenn.edu, Area de Conservacion Guanacaste, COSTA RICA, 11-SRNP-41294. Collecting date of caterpillar host: 20.iii.2011, collection date (eclosion date) of wasp: 05.iv.2011.

**Specimens examined.** Paratypes:  $1 \bigcirc , 1 \oslash (CNC)$  Costa Rica, same locality than holotype. Specimens voucher codes: DHJPAR0042995 and DHJPAR0043008.

**Description.** Promefur color: dark brown on anterior half, yellow on posterior half. Meso- and meta- femur color: fully dark brown to black (Fig. 4). Metatibia and metatarsus color: Metatibia with brown to black coloration on posterior 0.2–0.4 ×; metatarsus mostly dark brown, except for yellowish area on anterior half of first tarsomerus (Fig. 6). Tegula and humeral complex color: tegula and humeral complex fully yellow to yellowish-white (Fig. 9). Pterostigma color: centrally yellow-white, with thin brown margins (Fig. 5). Fore wing veins color: at least some veins with



Figures 4–9. *Iconella andydeansi*. 4 Habitus, lateral view 5 Fore wing 6 Ovipositor sheats and metatibia 7 Head, dorsal view 8 Propodeum 9 Meso- and metasoma (partially), dorsal view.

thin brown margins and interior of veins yellow to light brown. Body length (head to apex of metasoma): 2.7 mm or 2.8 mm. Fore wing length: 3.0 mm. Ocular-ocellar line/posterior ocellus diameter: 2.2 ×. Interocellar distance/posterior ocellus diameter:

2.1 × (Fig. 7). Antennal flagellomere 2 length/width: 2.5 × or 2.7 ×. Antennal flagellomere 14 length/width: 1.3 ×. Length of flagellomere 2/length of flagellomere 14: 2.4 ×. Metafemur length/width: 3.1 ×. Mesoscutellar disc: mostly with punctures scattered all over disc surface (Fig. 9). Number of pits in scutoscutellar sulcus: usually 12 or less, ocasionally reaching up to 14 pits. Propodeum background sculpture: anterior 0.2-0.4 × with rather dull puntures; posterior 0.6–0.8 × mostly sculptured, with mix of small puntures and carinae (mostly radiating from strong, longitudinal median carina) (Fig. 8). Mediotergite 1 width at anterior margin/width at posterior margin: 3.4 ×. Mediotergite 2 width at posterior margin/length: 4.4 × (Fig. 9). Ovipositor sheaths length/metatibial length: 1.1 × (Fig. 6).

Male. As female, although sculpture is slightly smoother.

Molecular data. We analyzed three full 658 bp barcodes for this species.

**Biology/ecology.** Host: An undescribed species of Phycitinae (Pyralidae) with provisional name "phyjanzen021 Janzen855" in the ACG database (http://janzen.sas. upenn.edu/caterpillars/database.lasso). Caterpillar collected while feeding on the foliage of *Lepidoploa salzmannii* (Asteraceae).

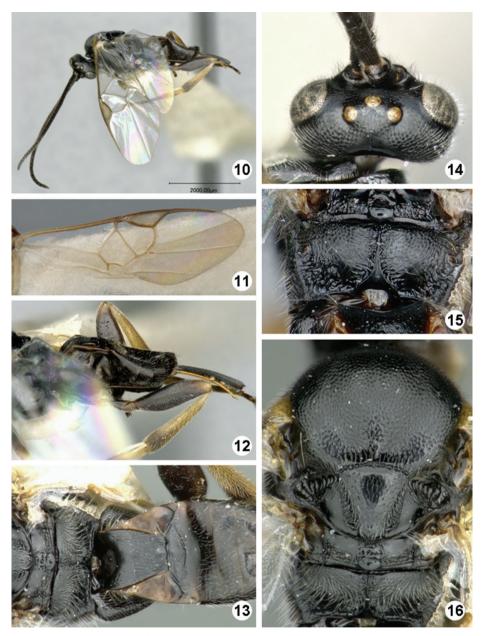
**Distribution.** Only known from the holotype locality, in Sector Rincon Rain Forest of ACG at 410 m.

**Comments.** The species has been reared only in one place from three caterpillars collected at the same time on the same species of food plant, out of 12,000+ rearings of ACG Pyralidae of more than 200 species. A single additional specimen, identified by DNA barcoding, has been reared in 2012 from the same species of host caterpillar in the same place and on the same food plant, but was not available for study. *Iconella andydeansi* is sympatric with *I. jayjayrodriguezae* in ACG, the latter being a species with a slightly larger ACG rain forest distribution but equally narrow food plant record, and also known from Chiapas, Mexico. Those are the only two species of New World *Iconella* that are known to be sympatric, and that was revealed through extensive collecting in ACG and the use of DNA barcodes. It is likely that further collecting in other areas of the Neotropics, as well as the barcoding of more fresh specimens, will reveal additional species.

**Etymology.** This species is named in honor of Andy R. Deans (Pennsylvania State University, United States) in recognition of his major contribution to the taxonomy of the many species in the microgatrine genus *Alphomelon* that occur in Area de Conservación Guanacaste (e.g., Deans et al. 2003).

*Iconella canadensis* Fernández-Triana, sp. n. http://zoobank.org/C3E8D164-ABBC-4823-9D56-50E302065F55 http://species-id.net/wiki/Iconella\_canadensis Figures 10–16

Type locality. CANADA. Ontario, Black Sturgeon Lake. Lat: 49.368333, Long: -88.881944.



Figures 10–16. *Iconella canadensis*. 10 Habitus, lateral view 11 Fore wing 12 Ovipositor sheats, meso-fermur, and metatibia 13 Propodeum, mediotergites 1–4, dorsal view 14 Head, dorsal view 15 Propodeum 16 Mesosoma, dorsal view.

**Holotype.**  $\bigcirc$ , CNC. First label: Black Sturgeon Lake, Ontario, Em. 1-6-viii-1961, Insectary. Second label: Nest 97, Cell 4 ex provisions. Third label: W61319. Fourth label: Host either A. betullela or Rh. hasta. Fifth label: DNA Voucher CNCHYM 01498.

**Specimens examined.** Paratypes:  $3 \ (CNC)$  Canada: ON, Black Sturgeon Lake, 21–29.vii.1961, 26.vii.1962, and 2.viii.1962, ex: Provisions Nests 52 and 66, one specimen with DNA Voucher CNCHYM01497;  $1 \ (CNC)$  Canada: ON, Galt, 11.vii.1952;  $1 \ (CNC)$  Canada: ON, Whitney, 4.vii.1949, ex: Phalaenidae;  $1 \ (CNC)$  Canada: NB, Kouchibouguac National Park, 30.viii.1967, code-6060B, DNA Voucher CNCHYM01511 and CNCHYM01512;  $1 \ (LFS)$  Canada: QC, Saint-Cléophas-de-Brandon, 4.vii.1968, ex: *Epinotia solandriana* on *Betula papyrifera*. Collecting dates of specimens examined: July and August (1949–1967).

Description. Promefur color: dark brown on anterior half, yellow on posterior half. Meso- and meta- femur color: mostly dark brown but with proximal  $0.1-0.2 \times$  yellow to orange (Fig. 12). Metatibia and metatarsus color: Metatibia with brown to black coloration on posterior 0.2–0.4 ×; metatarsus mostly dark brown, except for yellowish area on anterior half of first tarsomerus (Fig. 10). Tegula and humeral complex color: tegula and anterior half of humeral complex yellow to yellowish-white, posterior half of humeral complex light brown to dark brown. Pterostigma color: centrally yellow-white, with thin brown margins, rarely mostly brown, with yellowish-white spot at anterior  $0.2 \times$  (Fig. 11). Fore wing veins color: at least some veins with thin brown margins and interior of veins yellow to light brown. Body length (head to apex of metasoma): 3.5 mm, 3.6 mm, 3.7 mm, rarely 3.2 mm. Fore wing length: 3.8 mm, 3.9 mm, 4.0 mm, 4.1 mm or 4.2 mm. Ocular-ocellar line/posterior ocellus diameter: 1.6 ×. Interocellar distance/posterior ocellus diameter: 1.9 × (Fig. 14). Antennal flagellomere 2 length/width: 3.0 ×. Antennal flagellomere 14 length/width: 1.6 x. Length of flagellomere 2/length of flagellomere 14: 2.1 x. *Metafemur length/width:* 3.2 ×, 3.3 ×, rarely 3.4 ×. *Mesoscutellar disc:* mostly smooth with few, scattered punctures near margins (Fig. 16). Number of pits in scutoscutellar sulcus: usually 16, ocasionally only 14 pits. Propodeum background sculpture: anterior 0.2-0.4 × with rather dull puntures; posterior 0.6-0.8 × mostly sculptured, with mix of small puntures and carinae (mostly radiating from strong, longitudinal median carina) (Fig. 15). Mediotergite 1 width at anterior margin/width at posterior margin:  $2.1 \times \text{or} 2.2 \times .$ Mediotergite 2 width at posterior margin/length: 3.6 ×, 3.8 × or 4.4 × (Fig. 13). Ovipositor sheaths length/metatibial length:  $0.7 \times \text{or } 0.8 \times (\text{Fig. 12})$ .

Male. As female.

**Molecular data.** We analyzed three short 164 bp COI sequences from the DNA barcode region.

**Biology/ecology.** Host: *Epinotia solandriana* (Tortricidae) and likely *Acrobasis betulella* (Pyralidae) (see Comments below).

Distribution. Eastern Canada: NB, ON, QC.

**Comments.** The holotype has a label stating that it emerged from either *Acrobasis betulella* (Pyralidae) or *Rheumaptera hasta* (Geometridae). Based on the known biology of the genus *Iconella* in the world, the second alternative is unlikely, and thus we consider *A. betulella* as the potential host in that case. However, the pyralid host cannot be taken as definitive until more reared specimens confirm the decision.

**Etymology.** The name refers to the known distribution of the species, in Eastern Canada.

#### Iconella etiellae (Viereck, 1911)

http://species-id.net/wiki/Iconella\_etiellae Figures 17–22

*Apanteles etiellae:* Viereck 1911: 178. *Iconella etiellae* (Viereck). Mason 1981: 75.

# Type locality. UNITED STATES, Washington, Pullman.

**Holotype.** ♂, NMNH (examined).

**Specimens examined.**  $3 \ \bigcirc, 3 \ \Diamond$  (CNC) United States: CA (Apple Valley, Helendale, Kramer Hills and Panamint Valley) and UT (Dugway Proving Ground). Collecting dates of specimens examined: May and June (1953–1961).

Description. Promefur color: almost completely dark brown (yellow area absent or limited to posterior 0.2 x) (Fig. 17). Meso- and meta- femur color: mostly dark brown but with proximal 0.1-0.2 × yellow to orange (Fig. 19). Metatibia and metatarsus color: Metatibia mostly yellow, at most with very small and faint brown spot on posterior  $0.1 \times$  or less; metatarsus mostly yellow, except for brown area on posterior half of first tarsomerus (Fig. 19). Tegula and humeral complex color: tegula and anterior half of humeral complex yellow to yellowish-white, posterior half of humeral complex light brown to dark brown. Pterostigma color: mostly brown, with yellowish-white spot at anterior  $0.2 \times$  (Fig. 18). Fore wing veins color: most of veins transparent or at most yellowish-white, margins of same color than interior of vein. Body length (head to apex of metasoma): 3.0 mm or 3.1 mm. Fore wing length: 3.3 mm or 3.4 mm. Ocular-ocellar line/posterior ocellus diameter:  $1.6 \times \text{or} 1.7 \times \text{. Interocellar distance/posterior ocellus di$ ameter:  $2.4 \times \text{or} 2.6 \times (\text{Fig. 20})$ . Antennal flagellomere 2 length/width:  $2.6 \times \text{or} 2.7 \times .$ Antennal flagellomere 14 length/width: 1.4 × or 1.7 ×. Length of flagellomere 2/length of flagellomere 14: 2.2 ×. Metafemur length/width: 3.2 × or 3.3 ×. Mesoscutellar disc: mostly smooth with few, scattered punctures near margins (Fig. 22). Number of pits in scutoscutellar sulcus: usually 14 or more, ocasionally reaching up to 16 pits. Propodeum *background sculpture:* anterior  $0.2-0.4 \times$  with fine puntures; posterior  $0.6-0.8 \times$  mostly smooth, at most with some small carinae (mostly radiating from strong, longitudinal median carina) (Fig. 21). Mediotergite 1 width at anterior margin/width at posterior margin: 2.4 × or 2.5 ×. Mediotergite 2 width at posterior margin/length: 5.0 × or 5.1 × (Fig. 22). Ovipositor sheaths length/metatibial length:  $1.1 \times (Fig. 19)$ .

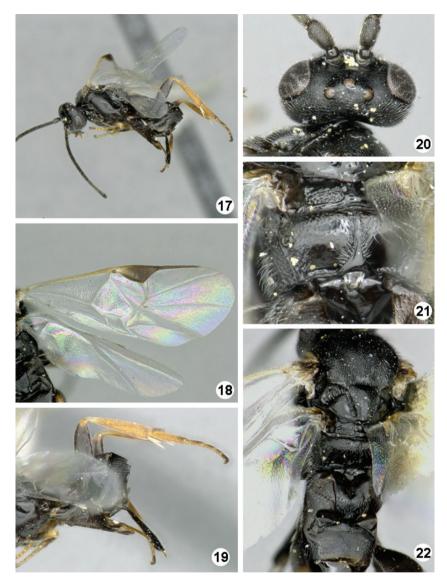
Male. As female.

Molecular data. We analyzed one 607 bp barcode for this species.

**Biology/ecology.** Hosts: *Etiella zinckenella, Olycella junctolineella, Psorosina ham*mondi, and Ufa rubedinella (Pyralidae).

Distribution. United States: AR, AZ, CA, CO, IA, NM, OK, WA.

**Comments.** There is a record from Virginia, United States (Yu et al. 2012). While we have not been able to examine that specimen, based in the known distribution of New World species of *Iconella* and their hosts, it is very likely that the VA specimen actually belongs to *Iconella canadensis* Fernández-Triana.

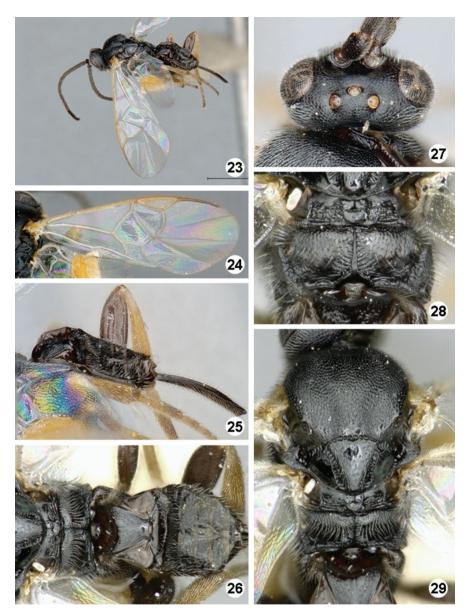


Figures 17–22. *Iconella etiellae*. 17 Habitus, lateral view 18 Fore wing 19 Ovipositor sheats, mesofermur, and metatibia 20 Head, dorsal view 21 Propodeum 22 Meso- and metasoma (partially), dorsal view.

*Iconella isolata* (Muesebeck, 1955), stat. r. http://species-id.net/wiki/Iconella\_isolata Figures 23–29

Apanteles etiellae isolatus Muesebeck, 1955: 161.

Type locality. TRINIDAD & TOBAGO, St. Augustine.



Figures 23–29. *Iconella isolata*. 23 Habitus, dorso-lateral view 24 Fore wing 25 Ovipositor sheats, mesofermur, and metatibia 26 Propodeum and metasoma, dorsal view 27 Head, dorsal view 28 Propodeum 29 Mesosoma and mediotergite I dorsal view.

Holotype. ♀, NMNH (not examined). Paratypes. 2 ♀ and 1 ♂, CNC (examined). Specimens examined. 11 ♀, 5 ♂ (CNC) British Guiana, Cayman islands (Grand Cayman, Georgetown), Grenada, Montserrat, Saint Kitts & Nevis (Nevis, Round Hill), Trinidad & Tobago (Paradise Mt., St. Augustine, and Tacarigua). Collecting dates of specimens examined: February, March and December (1950–1965).

Description. Promefur color: mostly yellow, dark brown area limited to anterior 0.2 or less, rarely dark brown on anterior half, yellow on posterior half. Meso- and meta- femur color: mostly dark brown but with proximal 0.1–0.2 × yellow to orange. Metatibia and metatarsus color: Metatibia mostly yellow, at most with very small and faint brown spot on posterior  $0.1 \times$  or less; metatarsus mostly yellow, except for brown area on posterior half of first tarsomerus. Tegula and humeral complex color: tegula and humeral complex fully yellow to yellowish-white (Fig. 29). Pterostigma color: centrally transparent, with yellow-white margins (Fig. 24). Fore wing veins color: most of veins transparent or at most vellowish-white, margins of same color than interior of vein. Body length (head to apex of metasoma): 2.4 mm, 2.5 mm, 2.6 mm, 2.7 mm, 2.8 mm or 2.9 mm. Fore wing length: 2.8 mm, 2.9 mm, 3.0 mm, 3.1 mm or 3.2 mm. Ocular-ocellar line/posterior ocellus diameter: 1.9 × or 2.0 ×. Interocellar distance/ posterior ocellus diameter: 1.9 ×, 2.0 × or 2.1 × (Fig. 27). Antennal flagellomere 2 length/width: 2.2 ×, 2.3 × or 2.5 ×. Antennal flagellomere 14 length/width: 1.3 × or 1.4 ×. Length of flagellomere 2/length of flagellomere 14: 2.3 ×, 2.4 × or 2.5 ×. Metafemur length/width: 3.2 × or 3.3 ×. Mesoscutellar disc: mostly smooth with few, scattered punctures near margins (Fig. 29). Number of pits in scutoscutellar sulcus: usually 12 or less, ocasionally reaching up to 14 pits. Propodeum background sculpture: anterior 0.2–0.4 × with fine puntures; posterior 0.6–0.8 × mostly smooth, at most with some small carinae (mostly radiating from strong, longitudinal median carina) (Fig. 28). Mediotergite 1 width at anterior margin/width at posterior margin: 2.1 ×, 2.6 × or 2.8 ×. Mediotergite 2 width at posterior margin/length: 3.6 ×, 3.7 ×, 3.8 ×, 4.5  $\times$  or 4.6  $\times$  (Fig. 26). Ovipositor sheaths length/metatibial length: 1.0  $\times$  (Fig. 25).

Male. As female.

**Molecular data.** No DNA barcode sequence was available for this species, the only one among the New World species without molecular data.

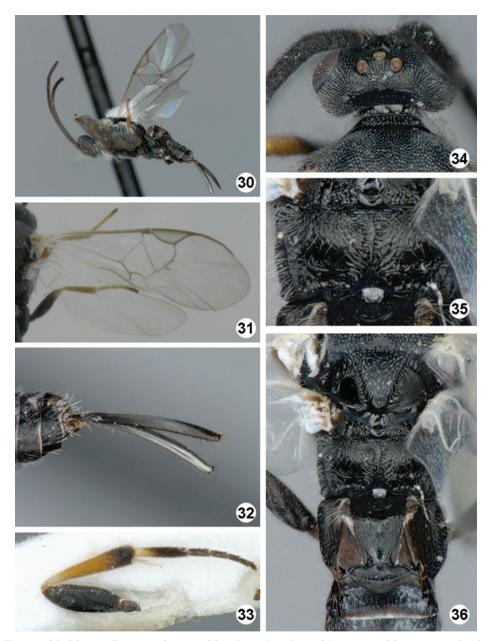
Biology/ecology. Host: Ancylostomia stercorea (Pyralidae).

**Distribution.** British Virgin Islands, Cayman Islands, Dominica, Grenada, Guyana, Montserrat, Puerto Rico, Saint Kitts & Nevis, Trinidad & Tobago.

**Comments.** Until now, *I. isolata* had been considered to be a subspecies of *I. etiellae*. After study of numerous specimens, we have found consistent and significant differences in morphology, hosts and geographical distribution and thus consider *I. isolata* as a different species on its own. Cayman Islands and Saint Kitts & Nevis are new locality records, based on CNC specimens. The specimen from Cayman Islands represents the westernmost locality, and it expands considerably the previously known distribution of the species in the Caribbean islands.

*Iconella jayjayrodriguezae* Fernández-Triana, sp. n. http://zoobank.org/A1E2E3FC-6C2D-4E6F-83C0-AD388D55DCB5 http://species-id.net/wiki/Iconella\_jayjayrodriguezae Figure 30–36

**Type locality.** COSTA RICA, Alajuela, Area de Conservacion Guanacaste, Sector Rincon Rain Forest, Sendero Venado, 420m. Lat: 10.89678, Long: -85.27001.



Figures 30–36. *Iconella jayjayrodriguezae*. 30 Habitus, lateral view 31 Fore wing 32 Ovipositor sheaths 33 Hind leg 34 Head, dorsal view 35 Propodeum 36 Meso- and metasoma (partially), dorsal view.

**Holotype.** ♀, NMNH. First label: DHJPAR0039740. Second label: Voucher: D.H.Janzen & W.Hallwachs, DB: http://janzen.sas.upenn.edu, Area de Conservacion Guanacaste, COSTA RICA, 09-SRNP-41791. Collecting date of caterpillar host 21.vii.2009, collection date (eclosion date) of wasp 10.viii.2009.

**Specimens examined.** Paratypes:  $3 \, \bigcirc, 1 \, \checkmark$  (CNC) Costa Rica, Alajuela, ACG, Sector San Cristobal, Rio Blanco Abajo, 500m, Lat: 10.90037 Long: -85.37254; 1  $\bigcirc$  (CNC) Mexico, Chiapas, 16°58'N, 91°47'W, 23–25.viii.1978. Collecting dates of specimens examined: January, March, April, June, July, and September (2009-2011) for Costa Rican specimens; August (1978) for Mexican specimen.

Description. Promefur color: mostly yellow, dark brown area limited to anterior 0.2 or less. Meso- and meta- femur color: mostly dark brown but with proximal 0.1-0.2 × yellow to orange. Metatibia and metatarsus color: Metatibia with brown to black coloration on posterior 0.2–0.4 ×; metatarsus mostly dark brown, except for yellowish area on anterior half of first tarsomerus (Fig. 33). Tegula and humeral complex color: tegula and humeral complex fully yellow to yellowish-white. *Pterostigma color:* centrally transparent, with yellow-white margins, rarely centrally yellow-white, with thin brown margins (Fig. 31). Fore wing veins color: at least some veins with thin brown margins and interior of veins yellow to light brown. Body length (head to apex of metasoma): 2.9 mm or 3.0 mm. Fore wing length: 3.2 mm or 3.3 mm. Ocular-ocellar line/posterior ocellus diameter: 2.0 ×. Interocellar distance/posterior ocellus diameter: 1.9 × (Fig. 34). Antennal flagellomere 2 length/width: 2.3 ×, 2.4 × or 2.5 ×. Antennal flagellomere 14 length/width: 1.3 ×, rarely  $1.1 \times \text{or} 1.5 \times$ . Length of flagellomere 2/length of flagellomere 14:  $2.3 \times, 2.4 \times, 2.5 \times \text{or} 2.6$ ×. Metafemur length/width: 3.2 ×, 3.3 × or 3.4 ×. Mesoscutellar disc: mostly smooth with few, scattered punctures near margins (Fig. 36). Number of pits in scutoscutellar sulcus: usually 12 or less, ocasionally reaching up to 14 pits. Propodeum background sculpture: anterior  $0.2-0.4 \times$  with rather dull puntures; posterior  $0.6-0.8 \times$  mostly sculptured, with mix of small puntures and carinae (mostly radiating from strong, longitudinal median carina) (Fig. 35). Mediotergite 1 width at anterior margin/width at posterior margin:  $3.1 \times 3.2 \times \text{or} 3.3 \times \text{.}$  Mediotergite 2 width at posterior margin/length:  $3.7 \times 3.8 \times 3.9$  $\times$  or 4.1  $\times$  (Fig. 36). Ovipositor sheaths length/metatibial length: 1.1  $\times$  (Fig. 32).

Male. As female.

**Molecular data.** We analyzed eight 650–658 bp barcodes for this species, one from Mexico and seven from Costa Rica.

**Biology/ecology.** Host: An undescribed species of Spilomelinae (Crambidae) with provisional name "spiloBioLep01 BioLep414" in the ACG database (http://janzen.sas. upenn.edu/caterpillars/database.lasso). Caterpillar collected while feeding on the foliage of *Lepidoploa salzmannii* and *Lepidoploa tortuosa* (Asteraceae).

**Distribution.** Costa Rica (ACG), and Mexico (Chiapas). In ACG it has been reared from four rainforest localities between 420–500m, 2-10 km apart, in six different months.

**Comments.** Although morphologically similar to *Iconella andydeansi*, *I. jayjayrod-riguezae* is known from two very widely separated places in Central America, and the barcoding sequences of the two species differ by 8.7% (57 bp).

**Etymology.** This species is named in honor of Josephine J. Rodriguez (National Center for Ecological Analysis and Synthesis, University of California, Santa Barbara, United States) in recognition of her outstanding enthusiasm for studying the taxonomy and biology of the microgastrine wasps of ACG (e.g., Smith et al. 2008).

#### Acknowledgements

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#### References

- Achterberg C van (2002) Western Palaearctic genera of the subfamily Microgastrinae: a reappraisal of the generic and tribal division (Hymenoptera: Braconidae). In: Melika G, Thuróczy G (Eds) Parasitic wasps: evolution, systematics, biodiversity and biological control. Agroinform, Budapest, Hungary, 19–35.
- Bartlett BR, Clausen CP, DeBach P, Goeden RD, Legner EF, McMurtry JA, Oatman ER (1978) Introduced parasites and predators of arthropod pests and weeds: a world review Agriculture Handbook No. 480, Agricultural Research Service, United States Department of Agriculture, Washington, District of Columbia.
- Bennett FD (1960) Parasites of Ancylostomia stercorea (Zell.) (Pyralidae, Lepidoptera), a pod borer attacking pigeon pea in Trinidad. Bulletin of Entomological Research 50: 737–757. doi: 10.1017/S0007485300054778
- Broad GR, Shaw MR, Godfray HCJ (2012) Checklist of British and Irish Braconidae (Hymenoptera). http://www.nhm.ac.uk/resources-rx/files/braconidae-checklist-final-34139.pdf [accessed 11.III.2013]
- Darriba D, Taboada GL, Doallo R, Posada D (2012) jModelTest 2: more models, new heuristics and parallel computing. Nature Methods 9: 772. doi: 10.1038/nmeth.2109
- Deans AR, Whitfield JB, Janzen DH (2003) Taxonomy and natural history of the microgastrine genus *Alphomelon* Mason (Hymenoptera: Braconidae). Journal of Hymenoptera Research 12(1): 1–41.
- Drummond AJ, Ashton B, Buxton S, Cheung M, Cooper A, Duran C, Field M, Heled J, Kearse M, Markowitz S, Moir R, Stones-Havas S, Sturrock S, Thierer T, Wilson A (2011) Geneious v6.0.5 http://www.geneious.com
- Fernández-Triana J (2010) Eight new species and an annotated checklist of Microgastrinae (Hymenoptera: Braconidae) from Canada and Alaska. ZooKeys 63: 1–53. doi: 10.3897/ zookeys.63.565
- Fernández-Triana J, Smith MA, Bodreault C, Goulet H, Hebert PDN, Smith AC, Roughley R (2011) A poorly known high-latitude parasitoid wasp community: unexpected diver-

sity and dramatic changes through time. PLoS ONE 6(8): e23719. doi: 10.1371/journal. pone.0023719

- Goloboff PA, Farris JS, Nixon KC (2008) TNT, a free program for phylogenetic analysis. Cladistics. 24: 774–786. doi: 10.1111/j.1096-0031.2008.00217.x
- Hajibabaei M, Smith MA, Janzen DH, Rodriguez JJ, Whitfield JB, Hebert PDN (2006) A minimalist barcode can identify a specimen whose DNA is degraded. Molecular Ecology Notes 6: 959–964. doi: 10.1111/j.1471-8286.2006.01470.x
- Huber JT, Sharkey MJ (1993) Structure. In: Goulet H, Huber JT (Eds) Hymenoptera of the world: an identification guide to families. Agriculture Canada Research Branch, Monograph No. 1894E, Ottawa, Canada, 13–59.
- Ivanova NV, Dewaard JR, Hebert PDN (2006) An inexpensive, automation-friendly protocol for recovering high-quality DNA. Molecular Ecology Notes 6 (4): 998–1002. doi: 10.1111/j.1471-8286.2006.01428.x
- Janzen DH, Hallwachs W, Blandin P, Burns JM, Cadiou J, Chacon I, Dapkey T, Deans AR, Epstein ME, Espinoza B, Franclemont JG, Haber WA, Hajibabaei M, Hall JPW, Hebert PDN, Gauld ID, Harvey DJ, Hausmann A, Kitching I, Lafontaine D, Landry J, Lemaire C, Miller JY, Miller JS, Miller L, Miller SE, Montero J, Munroe E, Rab Green S, Ratnasingham S, Rawlins JE, Robbins RK, Rodriguez JJ, Rougerie R, Sharkey MJ, Smith MA, Solis MA, Sullivan JB, Thiaucourt P, Wahl DB, Weller SJ, Whitfield JB, Willmott KR, Wood DM, Woodley NE, Wilson JJ (2009) Integration of DNA barcoding into an ongoing inventory of complex tropical biodiversity. Molecular Ecology Resources 9 (Supplement 1): 1–26. doi: 10.1111/j.1755-0998.2009.02628.x
- Mason WRM (1981) The polyphyletic nature of *Apanteles* Foerster (Hymenoptera: Braconidae): A phylogeny and reclassification of Microgastrinae. Memoirs of the Entomological Society of Canada 115: 1–147. doi: 10.4039/entm113115fv
- Meusnier I, Singer GAC, Landry JF, Hickey DA, Hebert PD, Hajibabaei M (2008) A universal DNA mini-barcode for biodiversity analysis. BMC Genomics 9: 214. doi: 10.1186/1471-2164-9-214
- Muesebeck CFW (1955) New reared Braconidae from Trinidad (Hymenoptera). Proceedings of the Entomological Society of Washington 57(4): 161–164.
- Nixon G (1965) A reclassification of the tribe Microgasterini (Hymenoptera: Braconidae). Bulletin of the British Museum (Natural History), Entomology series, Supplement 2: 1–284.
- Rambaut A, Drummond AJ (2009) Tracer v1.5. http://beast.bio.ed.ac.uk/Tracer
- Ronquist F, Huelsenbeck JP (2003) MRBAYES 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19: 1572–1574. doi: 10.1093/bioinformatics/btg180
- Saitou N, Nei M (1987) The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular Biology and Evolution 4: 406–425.
- Shorthouse DP (2010) SimpleMappr, an online tool to produce publication-quality point maps. http://www.simplemappr.net [accessed 13 March, 13]
- Smith MA, Woodley NE, Janzen DH, Hallwachs W, Hebert PDN (2006) DNA barcodes reveal cryptic host-specificity within the presumed polyphagous members of a genus of parasitoid flies (Diptera: Tachinidae). Proceedings of the National Academy of Sciences 103: 3657–3662. doi: 10.1073/pnas.0511318103

- Smith MA, Wood DM, Janzen DH, Hallwachs W, Hebert PDN (2007) DNA barcodes affirm that 16 species of apparently generalist tropical parasitoid flies (Diptera, Tachinidae) are not all generalists. Proceedings of the National Academy of Sciences 104: 4967–4972. doi: 10.1073/pnas.0700050104
- Smith MA, Rodriguez JJ, Whitfield JB, Deans AR, Janzen DH, Hallwachs W, Hebert PDN (2008) Extreme diversity of tropical parasitoid wasps exposed by iterative integration of natural history, DNA barcoding, morphology, and collections. Proceedings of the National Academy of Sciences 105: 12359–12364. doi: 10.1073/pnas.0805319105
- Smith MA, Fernández-Triana JL, Eveleigh E, Gómez J, Guclu C, Hallwachs W, Hebert PDN, Hrcek J, Huber JT, Janzen DH, Mason PG, Miller SE, Quicke DLJ, Rodriguez JJ, Rougerie R, Shaw MR, Varkonyi G, Ward D, Whitfield JB, Zaldívar-Riverón A (2013) DNA barcoding and the taxonomy of Microgastrinae wasps (Hymenoptera, Braconidae): impacts after 8 years and nearly 20 000 sequences. Molecular Ecology Resources, 13: 168– 176. doi: 10.1111/1755-0998.12038
- Stamatakis A (2006) RAxML-VI-HPC: maximum likelihotod-based phylogenetic analyses with housands of taxa and mixed models. Bioinformatics, 22(21): 2688–2690. doi: 10.1093/bioinformatics/btl446
- Tamura K, Nei M (1993) Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees. Molecular Biology and Evolution 10: 512–526.
- Viereck HL (1911) Descriptions of six new genera and thirty-one new species of Ichneumon flies. Proceedings of the United States National Museum 40(1812): 173–196.
- Whitfield JB (1997) Subfamily Microgastrinae. In: Wharton RA, Marsh PM, Sharkey MJ (Eds) Manual of the New World genera of the family Braconidae (Hymenoptera). Special Publication No. 1 International Society of Hymenopterists, Washington, D.C., 333–364.
- Yu DSK, van Achterberg C, Horstmann K (2012) Taxapad 2012, Ichneumonoidea 2011. Database on flash-drive. www.taxapad.com, Ottawa, Ontario, Canada.

# Appendix

Lucid key to the New World species of the parasitoid wasp *Iconella* (Hymenoptera, Braconidae, Microgastrinae). (doi: 10.3897/zookeys.321.5160.app) File format: Lucid Key Data (lk4).

**Copyright notice:** This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

**Citation:** Fernández-Triana JL, Cardinal S, Whitfield JB, Hallwachs W, Smith AM, Janzen DH (2013) A review of the New World species of the parasitoid wasp *Iconella* (Hymenoptera, Braconidae, Microgastrinae). ZooKeys 321: 65–87. doi: 10.3897/zookeys.321.5160 Lucid key to of the New World species of the parasitoid wasp *Iconella* (Hymenoptera, Braconidae, Microgastrinae). ZooKeys 321: 65–87. doi: 10.3897/zookeys.321.5160.app

SHORT COMMUNICATION



# First description of the female of Sinopoda serrata (Wang, 1990) (Araneae, Sparassidae)

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Citation: Quan D, Chen J, Jie Liu J (2013) First description of the female of *Sinopoda serrata* (Wang, 1990) (Araneae, Sparassidae). ZooKeys 321: 89–96. doi: 10.3897/zookeys.321.5752

#### Abstract

The female of *Sinopoda serrata* (Wang, 1990) is described for the first time from Tiantangzhai National Forest Park, Hubei province, China. This species has been recorded from the region of Central China. Morphological descriptions and illustrations of this species are given.

#### **Keywords**

Taxonomy, biodiversity, systematics, huntsman spiders

## Introduction

The spider genus *Sinopoda* Jäger, 1999 is distributed in East Asia and northern parts of South East Asia with 49 species described so far, 32 of which are known from China (Jäger 2012, Platnick 2013).

The species *Sinopoda serrata* (Wang, 1990) was first described in *Heteropoda* Latreille, 1804, based on male specimens only from Mt. Lushan (Jiangxi Province, China) and Mt. Huangshan (Anhui Province, China) (Wang 1990). Jäger (1999) transferred this species to the genus *Sinopoda*. Recently, the authors examined specimens collected from Hubei Province and found that 2 females and 7 males seemed to be this species based on comparison with the type specimens. Based on the currently known specimens, this species is distributed in the region of Central China (Fig. 17). The aim of the current paper is to re-describe the male and report the female for the first time, providing detailed morphological illustrations and photos.

#### Material and methods

Specimens were examined with an Olympus SZX16 stereomicroscope; details were further investigated with an Olympus BX51 compound microscope. All illustrations were made using an Olympus drawing tube. Male palp and epigyne were examined and illustrated after dissection from the spider bodies. Photos were made with a Canon G10 digital camera (14.7 megapixels) mounted on an Olympus SZX16 stereomicroscope. The digital images depicting the habitus and genital morphology were a composite of multiple images taken at different focal planes along the Z axis and assembled using the software package Helicon Focus 3.10. Most hairs and macrosetae were usually not depicted in the palp and epigyne drawings.

Leg measurements are shown as: total length (femur, patella, tibia, metatarsus, tarsus). Number of spines are listed for each segment in the following order: prolateral, dorsal, retrolateral, ventral (in femora and patellae ventral spines are absent and fourth digit is omitted in the spination formula).

Abbreviations: ALE—anterior lateral eyes, AME — anterior median eyes, C conductor, E — embolus, EA — embolic apophysis, FD — fertilization duct, GA — glandular appendage, LF — lateral furrow, LL — lateral lobes, LS — lobal septum, MSu — membranous sac unexpanded, PLE — posterior lateral eyes, PME — posterior median eyes, PP — posterior part of spermathecae, RTA — retrolateral tibial apophysis, T — tegulum. I, II, III, IV — legs I to IV. Collections: HBU — Hubei University, Wuhan, China; HNU — Hunan Normal University, Changsha, China.

#### Taxonomy

Family Sparassidae Bertkau, 1872 Subfamily Heteropodinae Thorell, 1873 Genus *Sinopoda* Jäger, 1999

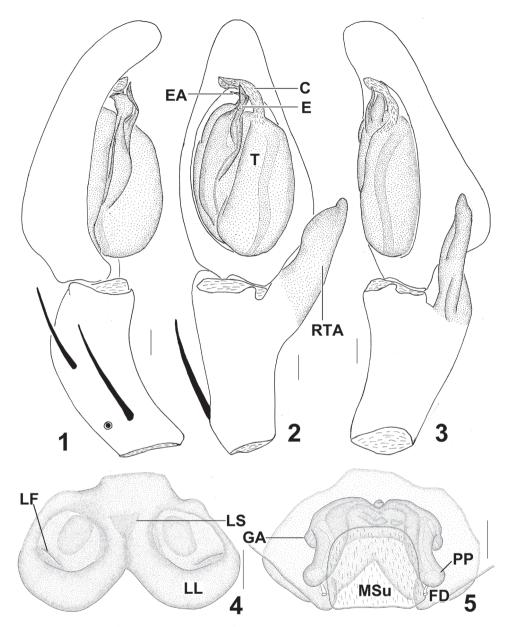
#### Sinopoda serrata (Wang, 1990)

http://species-id.net/wiki/Sinopoda\_serrata Figs 1–17

*Heteropoda serrata* Wang, 1990: 10, figs 17–19 (description of male). *Sinopoda serrata* Jäger, 1999: 21 (transferred from *Heteropoda*). *S. serrata* Song et al., 1999: 469, fig. 2700 (illustration of male).

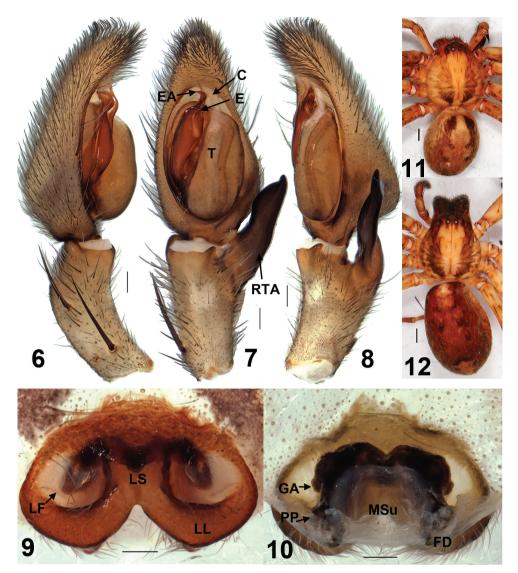
**Type material examined.** 1  $\Diamond$  (holotype, HNU), Mt. Lushan, Jiangxi Province, China, 15 June 1987, Xianjing Peng leg.; 1  $\Diamond$  (paratype, HNU), Mt. Huangshan, Anhui Province, China, October 1979, Jiafu Wang leg.

Additional material examined. 2  $3, 7 \ominus$  (HBU), Tiantangzhai National Forest Park (30°24'01.37"N, 115°18'19.31"E), Hubei, China, 8 September 2012, Fengxiang Liu, Jie Liu and Dan Quan leg.



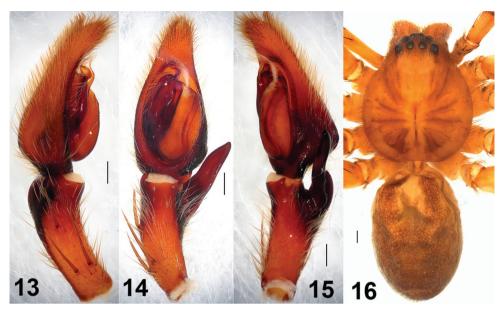
**Figures 1–5.** *Sinopoda serrata* (Wang, 1990), from Tiantangzhai National Forest Park (Hubei Province, China). I Left male palp, prolateral view **2** Left male palp, ventral view **3** Left male palp, retrolateral view **4** Epigyne, ventral view **5** Vulva, dorsal view. Scales = 0.2 mm. **C** conductor, **E** embolus, **EA** embolic apophysis, **FD** fertilization duct, **GA** glandular appendage, **LF** lateral furrow, **LL** lateral lobes, **LS** lobal septum, **MSu** membranous sac unexpanded, **RTA** retrolateral tibial apophysis, **PP**posterior part of spermathecae, **T** tegulum.

**Diagnosis.** Male of *S. serrata* is similar to *Sinopoda albofasciata* Jäger & Ono, 2002 in having the unbifurcated RTA, the slightly bent tip of embolus, but can be distinguished from the latter by the following characters: 1. RTA massive, but small



**Figures 6–12.** *Sinopoda serrata* (Wang, 1990), from Tiantangzhai National Forest Park (Hubei Province, China). **6** Left male palp, prolateral view **7** Left male palp, ventral view **8** Left male palp, retrolateral view **9** Epigyne, ventral view **10** Vulva, dorsal view **11** Male habitus, dorsal view **12** Female habitus, dorsal view. Scales = 0.2 mm (**6–10**), scales = 1 mm (**11–12**). **C** conductor, **E** embolus, **EA** embolic apophysis, **FD** fertilization duct, **GA** glandular appendage, **LF** lateral furrow, **LL** lateral lobes, **LS** lobal septum, **MSu** membranous sac unexpanded, **RTA** retrolateral tibial apophysis, **PP** posterior part of spermathecae, **T** tegulum.

in *S. albofasciata* (Jäger and Ono 2002); 2. The embolic apophysis (EA) distinctive, lamellar, but reduced in *S. albofasciata* (Jäger and Ono 2002) (Figs 1–3, 6–8). Female of *S. serrata* is similar to *Sinopoda undata* Liu, Li & Jäger, 2008 in having the fused lateral lobes (LL), the square-shaped membranous sac unexpanded (MSu), but can



**Figures 13–16.** *Sinopoda serrata* (Wang, 1990), holotype, from Mt. Lushan (Jiangxi Province, China). **13** Left male palp, prolateral view **14** Left male palp, ventral view **15** Left male palp, retrolateral view **16** Male habitus, dorsal view. Scales = 0.2 mm (**13–15**), scale = 1 mm (**16**).

be distinguished from the latter by the following characters: 1. Lobal septum (LS) of epigyne significantly short, but long in *S. undata* (Liu et al. 2008); 2. Internal duct system significantly wider than long, its left part widely separated from right part, but slightly wider than long in *S. undata*, its left part closed to right part (Liu et al. 2008) (Figs 4–5, 9–10).

**Description. Male:** Measurements: Prosoma length 4.97, width 4.19, anterior width 2.32, height 2.17; opisthosoma length 5.04, width 2.94. Eyes: AME 0.17, ALE 0.33, PME 0.23, PLE 0.29, AME–AME 0.24, AME–ALE 0.10, PME–PME 0.30, PME–PLE 0.37, AME–PME 0.41, ALE–PLE 0.36, clypeus height at AME 0.33, cl-ypeus height at ALE 0.28. Leg and palp measurements: Palp 7.33 (2.36, 1.34, 1.44, -, 2.19), I 20.38 (4.73, 1.97, 6.76, 5.04, 1.88), II 21.99 (5.96, 2.24, 5.61, 6.12, 2.06), III 16.22 (4.73, 1.63, 4.16, 4.10, 1.60), IV 18.16 (5.01, 1.73, 4.40, 5.03, 1.99). Leg formula: II-I-IV-III. Spination: palp 131, 101, 1021; femur I–III 323, IV 321; patella I–IV 101; tibia I–II, III 2126, IV 2326; metatarsus I–II 1014, III–IV 3036. Chelicerae yellowish-brown. Furrow with 3 anterior teeth, 4 or 6 posterior teeth, and with ca.80 denticles in elongated patch close to anterior teeth. Margins of fang base with one bristle. Palpal claw with 6 or 7 teeth. Sternum, ventral coxae and femora, distal legs as well as frontal chelicerae with long setae, otherwise with shorter setae.

Embolus (E) tip short, slender, slightly curved prolaterally, proximal part of embolus fully visible in the ventral view. Embolic apophysis (EA) short, lamellar, strongly



Figure 17. Collection localities of Sinopoda serrata (Wang, 1990) in China.

curved prolaterally. Sperm duct (SD) curved in ventral view. RTA large, not bifurcate, arising distally from tibia. Cymbium slightly longer than tibia (Figs 1–3, 6–8).

Colouration in ethanol (Fig. 11): Yellowish- to slightly yellowish-brown. Dorsal prosoma yellowish-brown with petaline patterns, which are divided by the bright yellowish region between posterior eye row and posterior margin of carapace. Sternum, ventral coxae and femora, gnathocoxae, and labium pale yellowish-brown, gnathocoxae and labium proximally reddish-brown. Chelicerae yellowish-brown. Legs pale yellowish-brown with distal parts slightly darker, dorsal femora with dark pattern. Dorsal opisthosoma with two pairs of spots situated in the median part, with a pale yellow inverted triangle-shaped pattern near the spinnerets. Lateral parts of opisthosoma reddish-brown, Ventral opisthosoma with little dark patterns.

**Female:** Measurements: Prosoma length 4.94, width 4.52, anterior width 2.49, height 2.15; opisthosoma length 7.33, width 4.86. Eyes: AME 0.22, ALE 0.31, PME 0.22, PLE 0.32, AME–AME 0.22, AME–ALE 0.11, PME–PME 0.32, PME–PLE 0.52, AME–PME 0.43, ALE–PLE 0.40, clypeus height at AME 0.32, clypeus height at ALE 0.33. Leg and palp measurements: Palp 6.23 (1.98, 0.94, 1.34, -, 1.97), I 14.37 (4.13, 1.82, 3.80, 3.51, 1.47), II 16.42 (4.61, 2.18, 4.26, 3.91, 1.46), III 13.42 (4.05, 1.68, 3.24, 3.15, 1.30), IV 15.64 (4.46, 1.54, 3.72, 4.16, 1.76). Leg formula: II-IV-I-III. Spination: palp 131, 100, 2121, 1014; femur I–III 323, IV 321; patella I 101, II–IV 001; tibia I–III 2026, IV 2326; metatarsus I–II 1014, III–IV 3036. Chelicerae yellowish-brown. Furrow with 3 anterior teeth, 4 posterior teeth, and with ca.80 denticles in elongated patch close to anterior teeth. Margins of fang base with one bristle. Palpal claw with 7 teeth. Sternum, ventral coxae and femora, distal legs as well as frontal chelicerae with long setae, otherwise with shorter setae.

Epigynal field wider than long. Lateral lobes (LL) fused, posteriorly with median incision. Epigynal pockets running from latero-posterior to medio-anterior, where copulatory openings are situated. Lateral furrows (LF) distinct, running to lateral margins of lateral lobes. Lobal septum (LS) wide, significantly short. Internal duct system significantly wider than long, its left part widely separated from right part. Glandular appendages (GA) small, extending not in posterior half of internal duct system. Posterior part of spermathecae (PP) strongly short, bulging slightly laterally. Fertilization ducts (FD) arising posterio-laterally. Membranous sac between fertilisation ducts unexpanded, almost square-shaped (Figs 4–5, 9–10).

Colouration in ethanol (Fig. 12) as in male.

**Remarks.** There is a small difference between the holotype male and the new collected materials: the middle part of RTA slightly covered the cymbium from the ventral view in the new collected materials, but not in the holotype (Figs 2, 7, 14).

Distribution. China (Hubei, Jiangxi, Anhui) (Fig. 17).

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### References

- Jäger P (1999) *Sinopoda*, a new genus of Heteropodinae (Araneae, Sparassidae) from Asia. The Journal of Arachnology, 27: 19–24.
- Jäger P, Ono H (2002) Sparassidae from Japan. II. First *Pseudopoda* species and new *Sinopoda* species (Araneae: Sparassidae). Acta Arachnology, Tokyo, 51: 109–124. doi: 10.2476/as-jaa.51.109
- Liu J, Li SQ, Jäger P (2008) New cave-dwelling huntsman spider species of the genus *Sinopoda* (Araneae: Sparassidae) from southern China. Zootaxa 1857: 1–20.
- Platnick NI (2013) The World Spider Catalog, Version 13.5. American Museum of Natural History. http://research.amnh.org/iz/spiders/catalog/index.html [accessed 1 June 2013]
- Song DX, Zhu MS, Chen J (1999) The Spiders of China. Hebei Science and Technology Publishing House, Shijiazhuang, 640 pp.
- Wang JF (1990) Six new species of the spiders of the genus *Heteropoda* from China (Araneae: Heteropodidae). Sichuan Journal of Zoology 9(3): 7–11.