RESEARCH ARTICLE



Discovery of Steninae from Ningxia, Northwest China (Coleoptera, Staphylinidae)

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Abstract

A study on the Steninae of Ningxia Autonomous Region is presented. Sixteen species are recognized, including new province records for 11 species and four new species: *Stenus biwenxuani* **sp. n.**, *S. liupanshanus* **sp. n.**, *Dianous yinziweii* **sp. n.**, *D. ningxiaensis* **sp. n.** Habitus photos of the new species, illustrations of diagnostic characters of all species and a key to species of the Steninae recorded from Ningxia are provided.

Keywords

Coleoptera, Staphylinidae, Steninae, China, Ningxia, identification key, new species

Introduction

Steninae, comprising two genera *Stenus* Latreille, 1797 and *Dianous* Leach, 1819, is a speciose subfamily of Staphylinidae. So far, 296 *Stenus* species and 103 *Dianous* species have been recorded from China. As far as the Steninae are concerned, Ningxia Autonomous Region is one of the most poorly explored regions, with merely two species recorded (Puthz 2008b): *Stenus deceptiosus* Puthz, 2008 and *Stenus comma* Leconte, 1863. In the summer of 2008, a team surveyed the insect fauna of the Liupan Shan Natural

Reserve in southern Ningxia and collected a large number of Steninae. In this paper, we report the results of the study on that material, which includes two new *Stenus* and two new *Dianous* species, and new province records for eleven *Stenus* species.

Material and methods

The specimens examined in this paper were collected by sifting leaf litter in forests and killed with ethyl acetate. For examination of the male genitalia, the last three abdominal segments were detached from the body after softening in hot water. The aedeagi, together with other dissected pieces, were mounted in Euparal (Chroma Gesellschaft Schmidt, Koengen, Germany) on plastic slides. Photos of sexual characters were taken with a Canon G7 camera attached to an Olympus SZX 16 stereoscope; habitus photos were taken with a Canon macro photo lens MP-E 65 mm attached to a Canon EOS40D camera.

Only records published after 2000 are given in the list of synonyms of each species. Articles published prior to 2001 may be found in Herman 2001.

The type specimens treated in this study are deposited in the following public and private collections:

cPut SHNU	private collection V. Puthz, Schlitz, Germany Department of Biology, Shanghai Normal University, P. R. China
The 1	neasurements of proportions are abbreviated as follows:
BL	body length, measured from the anterior margin of the clypeus to the posterior margin of abdominal tergite X
FL	forebody length, measured from the anterior margin of the clypeus to the apical margin of the elytra (apicolateral angle)
HW	width of head including eyes
PW	width of pronotum
EW	width of elytra
PL	length of pronotum
EL	length of elytra, measured from humeral angle

SL length of elytral suture

Taxonomy

Key to the species of Steninae of Ningxia

1	Labium unmodified; eyes relatively small, head mostly with distinct temples
	(Dianous)

_	Labium specialized, ejectable; eyes large, occupying entire lateral margin of
	head (<i>Stenus</i>)
2	First segment of metatarsus longer than the following segments combined;
	elytra without orange spots. Aedeagus: Fig. 24 D. inaequalis inaequalis
_	First segment of metatarsus not longer than the following segments com-
	bined; elytra each with an orange spot
3	Larger species, BL: 6.7mm; body with strong metallic luster, elytral spots
	larger and longitudinal. Sexual characters: Figs 49–52 D. ningxiaensis
_	Smaller species, BL: 4.8–5.1mm; body with faint metallic luster, elytral spots
	smaller and transverse. Sexual characters: Figs 42–48 D. yinziweii
4	Metatarsomere IV bilobed
_	Metatarsomere IV simple7
5	Larger species (BL: 5.5-6.7 mm), elytra with pair of orange spots. Sexual
	characters: Figs 9, 10 S. coronatus
_	Smaller species with BL less than 4.5 mm, elytra without spots6
6	Brachypterous species with elytra distinctly shorter than wide. BL: 2.6-
	2.7mm. Sexual characters: Figs 32-41 S. liupanshanus
_	Fully winged species with elytra longer than wide. BL: 3.2-4.0 mm. Sexual
	characters: Figs 11, 12 S. trigonuroides
7	Smaller species (BL: 2.5-3.1 mm), abdomen without paratergites. Sexual
	characters: Figs 13, 14 S. pilosiventris
_	Larger species with BL at least 3 mm, abdomen with paratergites
8	First three visible abdominal tergites with distinct basal keels
_	Abdominal tergites without basal keels
9	First three visible abdominal tergites with four basal keels. Aedeagus: Fig. 16
	S. melanarius melanarius
_	First three visible abdominal tergites with three basal keels
10	Smaller species with elytra distinctly shorter than wide. BL: 3.0–3.2mm. Ae-
	deagus: Fig. 15
_	Larger species (BL at least 4.4 mm) with elvtra longer than wide
11	Smaller species with reddish legs. BL: 4.4–4.7mm. Aedeagus: Fig. 18
_	Larger species with black legs, BL: 5.4–6.0 mm, Aedeagus: Fig. 17 S. juno
12	Elvtra without orange spots. BL: 5.4 mm. Sexual characters: Figs 26–30
	S. biwenxuani
_	Elvtra with pair of orange spots
13	Legs reddish. BL: 4.8–5.2 mm. Aedeagus: Fig. 19 S. alienus
_	Legs black
14	Smaller species, BL: 3.5–4.3 mm, elvtral punctation extremely dense. Aedea-
	gus: Fig. 20
_	Larger species with BL at least 4.3 mm. elvtral punctation less dense
15	Lateral portions of frons sparsely punctate. interstices at least as wide as diam-
-	eter of punctures. BL: 4.4–5.6 mm. Aedeagus: Fig. 21

-	Lateral portions of frons densely punctate, interstices smaller than diameter
	of punctures16
16	Male paratergites with punctures arranged in two irregular rows. BL: 4.3–5.5
	mm. Aedeagus: Figs 18, 20, 21 in Puthz 2008 S. comma
_	Male paratergites with punctures mostly arranged in one irregular row. BL:
	4.4-5.5 mm. Aedeagus: Figs 22, 23 S. falsator

Stenus coronatus Benick, 1928

http://species-id.net/wiki/Stenus_coronatus Figs 9, 10

Distribution. China (Ningxia, Yunnan, Shanxi, Henan, Hebei, Beijing, Jilin), Korea, Japan.

Stenus trigonuroides Zheng, 1993

http://species-id.net/wiki/Stenus_trigonuroides Figs 11, 12

Stenus trigonuroides Zheng, 1993: 229; Puthz 2008a: 173.

Material examined: China: Ningxia: $2 \Im \Im$, $2 \Im \Im$, Jinyuan County, Fengtai Linchang, 2300 m, 27–28.VI.2008, Wen-Xuan Bi leg.

Distribution. China (Ningxia, Sichuan, Liaoning).

Stenus pilosiventris Bernhauer, 1915

http://species-id.net/wiki/Stenus_pilosiventris Figs 13, 14

Material examined: China: Ningxia: 2 3, 1 2, Jinyuan County, Sutai Linchang, 2300m,21.VI.2008, Zi-Wei Yin leg.

Distribution. China (Ningxia, Shanxi, Shanghai, Beijing), Korea, Mogolia, Russia.

Stenus puthzi Hromádka, 1977

http://species-id.net/wiki/Stenus_puthzi Fig. 15

Stenus puthzi Hromádka, 1977: 7. Stenus asprohumilis Zhao & Zhou, 2006: 284; Puthz 2008a: 151. Distribution. China (Ningxia, Shanxi, Heilongjiang), Russia.

Stenus melanarius melanarius Stephens, 1833

http://species-id.net/wiki/Stenus_melanarius_melanarius Fig. 16

Material examined: China: Ningxia: 1 ♂, 2 ♀♀, Longde County, Sutai Linchang, 2200 m, 22.VI.2008, Zi-Wei Yin leg.

Distribution. Widely distributed in the Palaearctic region.

Stenus juno Paykull, 1789

http://species-id.net/wiki/Stenus_juno Fig. 17

Material examined: China: Ningxia: 1 ♀, Jinyuan County, Fengtai Linchang, 2300 m, 27–28. VI.2008, Wen-Xuan Bi leg.

Distribution. Widely distributed in the Holarctic region.

Stenus secretus Bernhauer, 1915

http://species-id.net/wiki/Stenus_secretus Fig. 18

Material examined: China: Ningxia: 1 ♂, Jinyuan County, Guamagou Linchang, 2200 m, 4.VII.2008, Wen-Xuan Bi leg.

Distribution. China (Ningxia, Heilongjiang, Shanxi), Korea, Russia.

Stenus alienus Sharp, 1874

http://species-id.net/wiki/Stenus_alienus Fig. 19

Stenus alienus Sharp, 1874: 81; Puthz 2008b: 176.

Material examined: China: Ningxia: 1 ♂, Jinyuan County, Qiuqianjia, 1800 m, 6-VI-2008, Wen-Xuan Bi leg.

Distribution. China (Ningxia, Qinghai, Shaanxi, Shanxi, Beijing, Taiwan), Russia, Mogolia, Korea, Japan.

Stenus scabratus Puthz, 2008

http://species-id.net/wiki/Stenus_scabratus Fig. 20

Stenus scabratus Puthz, 2008b: 180.

Material examined: China: Ningxia: 3 ♂♂, 1 ♀, Jinyuan County, Hongxia Lingchang, 2000 m, 11–12.VI.2008, Wen-Xuan Bi leg. Distribution. China (Ningxia, Sichuan, Yunnan)

Stenus deceptiosus Puthz, 2008

http://species-id.net/wiki/Stenus_deceptiosus Fig. 21

Stenus deceptiosus Puthz, 2008b: 184.

Material examined: China: Ningxia: 7 \Im , 7 \Im , 9 \Im , Jinyuan County, Qiuqianjia, 1800 m, 6.VII.2008, Wen-Xuan Bi leg.; 2 \Im , 3 \Im , 9 \Im , Jingyuan County, Xixia, 15.VII. 2008, Feng Yuan leg.

Distribution. China (Shaanxi, Shanxi, Ningxia, Hebei, Bejing, Liaoning), Korea.

Stenus falsator Puthz, 2008

http://species-id.net/wiki/Stenus_falsator Figs 22, 23

Stenus falsator Puthz, 2008b: 182.

Material examined: China: Ningxia: 8 \Im , 7 \Im , Jinyuan County, Qiuqianjia, 1800 m, 6.VII.2008, Wen-Xuan Bi leg.; 7 \Im , 8 \Im , 9 \Im , Jingyuan County, Xixia, 15.VII. 2008, Feng Yuan leg.

Distribution. China (Shanxi, Beijing, Jilin, Heilongjiang, Neimengu), Russia.

Stenus biwenxuani sp. n. urn:lsid:zoobank.org:act:26EF1235-E3F4-427D-A3F8-31DFF08BFB05 http://species-id.net/wiki/Stenus_biwenxuani Figs 1, 2, 26–30

Type material. Holotype. China: Ningxia: ∂, glued on a card with labels as follows: "Jinyuan County, Erlonghe Linchang, 2100 m, 9.VII.2008, Wen-Xuan Bi leg." "Holotype / *Stenus biwenxuani* / Tang & Li" [red handwritten label] (SHNU).



Figures 1-4. Habitus of Stenus. 1, 2 S. biwenxuani 3, 4 S. liupanshanus. Scales = 1 mm.

Diagnosis. The new species belongs to the *Stenus comma* group, and is similar to *Stenus atrovestis* Puthz, 2008 (Puthz 2008b). However it can be easily distinguished from the latter by the reddish brown legs, longer elytra and simple metatibiae (*S. atrovestis* with black legs, shorter elytra and flattened metatibiae).

Description. Body blackish with a faint plumbeous luster, antennae dark brown with club darker, maxillary palpi yellowish with last and apical half of penultimate segments brownish, legs reddish brown except knee darker with a faint plumbeous luster.

BL: 5.4 mm; FL: 2.7 mm.

HW: 1.00 mm, PL: 0.85 mm, PW: 0.80 mm, EL: 1.15 mm, EW: 1.11 mm, SL: 0.93 mm.

Head 0.90 times as wide as elytra; interocular area with deep longitudinal furrows, median portion moderately convex, not reaching the level of inner eye margins; punctures round, extremely dense, and of similar size; diameter of punctures about as wide as apical cross section of antennal segment III; interstices much narrower than half the diameter of punctures except those along the midline of the convex median portion, which may be a little broader than half the diameter of punctures. Antennae, when reflexed, extending a little beyond middle of pronotum; relative length of antennal segments from base to apex as 12 : 10 : 17.5 : 10: 9 : 6.5 : 7: 5 : 6 : 6 : 10. Paraglossa oval.

Pronotum 1.06 times as long as wide; disc with shallow and broad median longitudinal furrow fused with pairs of shallow impressions in anterior half, in the middle, and in posterior half; punctures round and very dense, slightly confluent, a little larger than those of head; interstices partially reticulated, of variable width, as wide as half the diameter of punctures or narrower.

Elytra 1.04 times as long as wide; disc slightly uneven with indistinct longitudinal humeral impression, indistinct postero-lateral impression, and indistinct sutural impression; punctures mostly confluent, a little larger than those of pronotum with rugose interstices.

Hind tarsi 0.76 times as long as hind tibiae, tarsomeres IV simple.

Abdomen semi-cylindrical with broad, raised and densely punctate paratergites of segments III–VI, width of paratergites of segment III slightly broader than apical width of metatibiae, punctures slightly larger than those on median portion of tergites; posterior margin of tergite VII with palisade fringe; punctures on abdominal tergites III–VIII round to elliptic, very dense, gradually becoming smaller posteriad; interstices mostly as wide as half the diameter of punctures at most, with relatively faint reticulation on all abdominal tergites.

Male. Mesotibiae and metatibiae each with a subapical tooth on inner side; sternite VI impressed postero-medially with a shallow emargination along the posterior margin of the impression; sternite VII impressed medially, posterior margin of this impression emarginate; sternite VIII (Fig. 26) with emargination at middle of posterior margin; sternite IX (Fig. 27) with apico-lateral projections long and stout, posterior margin serrate; tergite X (Fig. 28) with posterior margin slightly emarginated. Aedeagus (Figs

29, 30) slender, median lobe with a very long and pointed apex; internal plate strongly sclerotized (Fig. 31), parameres extending beneath apex of median lobe, widened and folded in apical third, each with 18 setae on inner side.

Female. unknown.

Distribution. China (Ningxia).

Etymology. This species is named in honor of Mr. Wen-Xuan Bi, the collector of the new species.

Stenus liupanshanus sp. n.

urn:lsid:zoobank.org:act:3AA00430-6919-4876-B0DD-AAC1926A26C3 http://species-id.net/wiki/Stenus_liupanshanus Figs 3, 4, 32–41

Type material. Holotype. China: Ningxia: 3, glued on a card with labels as follows: "Jinyuan County, Fengtai Linchang, 2400 m, 26.VI.2008, Wen-Xuan Bi & Zi-Wei Yin leg." "Holotype / *Stenus liupanshanus* / Tang & Li" [red handwritten label] (SHNU). **Paratypes.** 1 9, same data as for the holotype (SHNU); 1 3: ibidem, 2310m, 22.VI.2008, idem (cPut); 2 33, Jinyuan County, Dongshanpo, 2310 m, 27.VI.2008, Wen-Xuan Bi leg. (SHNU); 2 99, Jinyuan County, Heshangpu Linchang, 2300 m, 27.VI.2008, Wen-Xuan Bi leg. (SHNU); 2 99, Jinyuan County, Qiuqianjia, 1800 m, 6.VII.2008, Wen-Xuan Bi leg. (SHNU)

Diagnosis. The new species belongs to the *Stenus cephalotes* group and can easily be distinguished from other Chinese representatives of this group by the presence of distinct reticulation on the forebody and the very short elytra (EL/EW less than 0.8).

Description. Brachypterous; body dark brown with head slightly darker, anterior margin of labrum, antennae, maxillary palpi and legs reddish brown.

BL: 2.6–2.7mm; FL: 1.3 mm.

HW: 0.62–0.67 mm, PL: 0.44–0.48 mm, PW: 0.49–0.52 mm, EL: 0.45–0.51 mm, EW: 0.60–0.64 mm, SL: 0.32–0.34 mm.

Head 1.02–1.04 times as wide as elytra; interocular area with deep longitudinal furrows, median portion convex, slightly extending beyond the level of inner eye margins; punctures round, moderately confluent, and of similar size, diameter of punctures about as wide as apical cross section of antennal segment III; interstices rugose with indistinct reticulation, much narrower than half the diameter of punctures except those along the midline of the convex median portion, where they may be slightly broader than diameter of punctures. Antennae, when reflexed, not reaching middle of pronotum; relative length of antennal segments from base to apex as 6: 5.5: 8: 5: 5: 4: 3: 2.5: 4: 4: 5.5. Paraglossa oval.

Pronotum 0.91–0.93 times as long as wide; disk somewhat flattened, with shallow median longitudinal furrow; punctures slightly confluent, a little larger than those of head; interstices reticulated, distinctly narrower than half the diameter of punctures.

Elytra 0.75–0.79 times as long as wide, distinctly constricted at base; lateral margins gently divergent posteriad; disk rather even, suture slightly convex; punctation and interstices similar to those of pronotum.

Legs with hind tarsi 0.68 times as long as hind tibiae, tarsomeres IV distinctly bilobed.

Abdomen cylindrical; distinct paratergites absent, but rudimentary lateral border present; posterior margin of tergite VII with palisade fringe; punctures of abdominal tergites III–VIII elliptic, gradually becoming smaller posteriad; interstices narrower than half the diameter of punctures, with relatively faint microsculpture on tergites III–VII and distinct reticulation on tergites VIII–X.

Male. Sternite VIII (Fig. 32) with very shallow emargination at middle of posterior margin; sternite IX (Fig. 33) with apicolateral projections very long and posterior margin serrate and emarginate; tergite X (Fig. 34) with posterior margin convex. Aedeagus (Figs 35, 36) with median lobe roundly pointed at apex; expulsion hooks (Fig. 37) very large; parameres extending a little beyond apex of median lobe, dilated in apical third, each with two groups of setae on inner side: 5–6 apical setae and 5–6 subapical setae.

Female. Abdomen broader than that of male; sternite VIII (Fig. 38) slightly produced in the middle of posterior margin; tergite X (Fig. 39) similar to that of male; sclerotized spermatheca bent twice with many bubble structures on second tube (Figs 40, 41).

Distribution. China (Ningxia).

Etymology. The specific name is derived from "Liupanshan", the mountain where the type specimens were found.

Dianous inaequalis inaequalis Champion, 1919

http://species-id.net/wiki/Dianous_inaequalis_inaequalis Fig. 24

Dianous inaequalis Champion, 1919: 45. *Dianous caeruleoguttatus* Cameron, 1927: 6, 8.

Material examined: China: Ningxia: 1 \mathcal{J} , 2 $\mathcal{Q}\mathcal{Q}$, Jinyuan County, Qiuqianjia, 1800 m, 6.VI.2008, Wen-Xuan Bi leg.

Distribution. China (Yunnan, Sichuan, Ningxia), India.

Dianous yinziweii sp. n.

urn:lsid:zoobank.org:act:DC4E723C-CE73-4E38-88B1-168948840CD8 http://species-id.net/wiki/Dianous_yinziweii Figs 5, 6, 42–48

Type material. Holotype. China: Ningxia: ♂, glued on a card with labels as follows: "Jinyuan County, Erlonghe Linchang, Xiaonanchuan, 2000 m, 10.VII.2008, Zi-Wei



Figures 5-8. Habitus of *Dianous.* 5, 6 *D. yinziweii* 7, 8 *D. ningxiaensis.* Scales = 1 mm.

Yin leg." "Holotype / *Dianous yinziweii* / Tang & Li" [red handwritten label] (SHNU). **Paratypes.** 155 ♂♂, 129 ♀♀, same data as for the holotype (2 pair in cPut, remainder in SHNU); 18 ♂♂, 28 ♀♀, Jingyuan County, Erlonghe Linchang, 2200 m, 22.VII. 2008, Feng Yuan leg. (SHNU); 1 ♂, Jinyuan County, Fengtai Linchang, 2400 m, 26.VI.2008, Wen-Xuan Bi leg. (SHNU)

Diagnosis. The new species belongs to the *Dianous chinensis* complex and is similar to *Dianous banghaasi* Bernhauer, 1915 in sharing the elytral spots reaching the lateral margins in dorsal view. However, it can be easily distinguished from the latter by the distinctly smaller body size and the faint metallic luster of the entire body, which is strongly metallic blue in *D. banghaasi*.

Description. Body black with a plumbeous luster, antennal club brownish, elytra each with a large transverse orange spot, which reaches the lateral margins of the elytra in dorsal view, and with a narrow band of coppery luster around the spot, pubescence silvery to golden brown throughout, that of elytral spots golden brown.

BL: 4.8–5.1mm; FL: 2.5–2.8 mm.

HW: 0.98–1.04 mm, PL: 0.83–0.85 mm, PW: 0.77–0.82 mm, EL: 1.17–1.22 mm, EW: 1.07–1.16 mm, SL: 0.98–1.00 mm

Head 0.85–0.94 times as wide as elytra; interocular area with deep longitudinal furrows, median portion convex; punctures round, slightly confluent along the furrows, larger and sparser in median area than those near inner margins of eyes, diameter of large punctures about as wide as apical cross section of antennal segment III; interstices smooth, much narrower than half the diameter of punctures. Antennae, when reflexed, extending distinctly beyond posterior margin of pronotum; relative length of antennal segments from base to apex as 13.5: 9: 20: 14: 13.5:12: 11.5: 11: 11: 10: 12.5.

Pronotum 1.03–1.08 times as long as wide; disk relatively even; punctures round, transversely confluent in posterior portion, a little larger than those on head; interstices smooth, narrower than half the diameter of punctures except those in median portion, which may be as broad as two or three punctures.

Elytra 1.03–1.09 times as long as wide; punctation and interstices similar to those of pronotum, except that punctation of basal half portion and along suture is distinctly confluent with rugose interstices.

Hind tarsi with tarsomeres IV distinctly bilobed.

Abdomen semi-cylindrical with broad, raised and densely punctate paratergites of segments III–VI, width of paratergites of segment III as broad as apical width of metatibiae, punctures minute; posterior margin of tergite VII with palisade fringe; punctures on abdominal tergites III–VIII minute, smaller than ommatidia of eyes; interstices without microreticulation except tergite VIII, varied from a little narrower than half the diameter of punctures to much broader than diameter of punctures.

Pubescence of fore body long and suberect, single setae as long as fourth antennal segment.

Male. Sternite VII impressed postero-medially with shallow emargination along posterior margin of the impression; sternite VIII (Fig. 42) with deep emargination in the middle of posterior margin; sternite IX (Fig. 43) with apicolateral projections mod-



Figures 9–16. 9, 11, 13, 15, 16 Aedeagi of *Stenus*. 10, 12, 14 Spermathecae of *Stenus* 9, 10 *S. coronatus* 11, 12 *S. trigonuroides* 13, 14 *S. pilosiventris* 15 *S. puthzi* 16 *S. melanarius melanarius*. Scales = 0.25 mm.

erately pointed and posterior margin serrate; tergite X (Fig. 44) with posterior margin slightly emarginated. Aedeagus (Fig. 45) with median lobe bilobed at apex; parameres slightly bent inwards, extending distinctly beyond the apex of median lobe, with setae on inner side of apical portion.

Female. Abdomen slightly broader than that of male; sternite VIII (Fig. 46) distinctly produced in the middle of posterior margin; valvifer (Fig. 47) with posterior margin finely serrate; tergite X (Fig. 48) with posterior margin convex.

Distribution. China (Ningxia).

Etymology. This species is named in honor of Mr. Zi-Wei Yin, the collector of the new species.



Figures 17–25. 17–22, 24, 25 Aedeagi of *Stenus* and *Dianous* 23 Internal plate of aedeagus of *Stenus*. 17 *S. juno* 18 *S. secretus* 19 *S. alienus* 20 *S. scabratus* 21 *S. deceptiosus* 22, 23 *S. falsator* 24 *D. inaequalis inaequalis* 25 *D. chinensis.* Scales = 0.25 mm.

Dianous ningxiaensis sp. n.

urn:lsid:zoobank.org:act:25C5CED3-6704-4985-93FB-8E2FBB45C32E http://species-id.net/wiki/Dianous_ningxiaensis Figs 7, 8, 49–52

Type material. Holotype. China: Ningxia: (3), glued on a card with labels as follows: "Jinyuan County, Erlonghe Linchang, Xiaonanchuan, 2000 m, 10.VII.2008, Zi-Wei Yin leg." "Holotype / *Dianous ningxiaensis* / Tang & Li" [red handwritten label] (SHNU).



Figures 26–31. *Stenus biwenxuani*. 26 male sternite VIII 27 male sternite IX 28 male tergites IX, X 29, 30 aedeagus 31 sclerotized plate of aedeagus. Scales = 0.25 mm.

Diagnosis. The new species belongs to the *Dianous chinensis* complex and is similar to *Dianous chinensis* Bernhauer, 1915 (Fig. 25). It can be easily distinguished from the latter by the extremely large elytral spots and distinctly longer parameters of the aedeagus.

Description. Body black with a blue to purple metallic luster, antennal club brownish, each elytron with a large elongate orange spot, which is 1/2 as long as and



Figures 32–41. *Stenus liupanshanus.* **32** male sternite VIII **33** male sternite IX **34** male tergites IX, X **35, 26** aedeagus **37** expulsion hooks **38** female sternite VIII **39** female tergites IX, X **40** valvifers and spermatheca **41** spermatheca. Scales = 0.25 mm.



Figures 42–48. *Dianous yinziweii*. 42 male sternite VIII 43 male sternite IX 44 male tergites IX, X 45 aedeagus 46 female sternite VIII 47 valvifers 48 female tergites IX, X. Scales = 0.25 mm.

3/5 as broad as the respective elytron, and with a coppery luster around the spot, pubescence silvery to golden brown throughout, that of elytral spots golden brown.

BL: 6.7 mm; FL: 3.3 mm.

HW: 1.07 mm, PL: 0.91 mm, PW: 0.87 mm, EL: 1.55 mm, EW: 1.40 mm, SL: 1.28 mm.



Figures 49–52. *Dianous ningxiaensis*. 49 male sternite VIII 50 male sternite IX 51 male tergites IX, X 52 aedeagus. Scales = 0.25 mm.

Head 0.77 times as wide as elytra; interocular area with deep longitudinal furrows, median portion convex; punctures round, similar in size, diameter of punctures about as wide as basal cross section of antennal segment III; interstices without microsculpture and of variable width, ranging from being narrower than half the diameter of punctures to being of similar width as diameter of punctures. Antennae, when reflexed, extending distinctly beyond posterior margin of pronotum; relative length of antennal segments from base to apex as 16: 9.5: 34.5: 16.5: 18: 15.5: 15: 14: 12: 13.

Pronotum 1.04 times as long as wide; disk uneven, with two deep median impressions fused with a distinct basal impression; punctures mostly well delimited, slightly larger than those on head; interstices without microreticulation and of variable width.

Elytra 1.11 times as long as wide; punctation and interstices similar to those of pronotum except for a few larger punctures and partly fainly microsculptured interstices.

Hind tarsi with tarsomeres IV distinctly bilobed.

Abdomen semi-cylindrical with broad, raised and densely punctate paratergites of segments III–VI, paratergites of segment III slightly broader than apical width of metatibiae, punctures minute; posterior margin of tergite VII with palisade fringe; punctures on abdominal tergites III–VIII minute, smaller than ommatidia of eyes; interstices without microsculpture, except those of sternite VIII and of variable width.

Pubescence of fore body conspicuously long and suberect, single setae as long as fourth antennal segment.

Male. Sternite VII with posteromedian portion slightly flattened and densely pubescent; sternite VIII (Fig. 49) with deep emargination in the middle of posterior margin; sternite IX (Fig. 50) with apicolateral projections moderately pointed and posterior margin serrate; tergite X (Fig. 51) with posterior margin slightly emarginated. Aedeagus (Fig. 52) with median lobe bilobed at apex; parameres bent inwards, extending distinctly beyond apex of median lobe, with setae on inner side of apical portion.

Female. Unknown.

Distribution. China (Ningxia).

Etymology. The specific name is derived from "Ningxia", the type locality of this species.

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RESEARCH ARTICLE



A new species of Myrmozercon Berlese (Acari, Mesostigmata, Laelapidae) associated with ant from Iran

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Abstract

This paper report on a new species of mites of the genus Myrmozercon associated with ant in Iran – Myrmozercon cyrusi Ghafarian and Joharchi **sp. n.** was collected associated of the *Monomorium* sp. in Kenevist Rural District in the Central District of Mashhad County, Khorasan Razavi Province, Iran. This new species is described and illustrations provided. *Myrmozercon ovatum* Karawajew, 1909 is suspected to be a junior synonym of *M. brevipes* Berlese, 1902 and host-specificity and host range of *Myrmozercon* are also reviewed.

Keywords

Laelapidae, Myrmozercon, ants, taxonomy, Iran, myrmecophiles, nest

Introduction

Many species Laelapidae have been reported from ants or their nests. The myrmecophiles genus *Myrmozercon* includes about 22 described species from Europe, Australia, Africa, Middle East, Transcaucasia, North America and Central Asia (Michael 1891,

Hunter and Hunter 1963, Rosario and Hunter 1988, Karawajew 1909, Ueckermann and Loots 1995, Walter 2003, Shaw and Seeman 2009, Trach and Khaustov 2011, Joharchi et al. 2011).

All species are associated with ants, except for one intercepted at quarantine on plant material (Hunter and Hunter 1963). Shaw and Seeman (2009) synonymised *Parabisternalis* Ueckermann and Loots, 1995 with *Myrmozercon*, and included the subgenus *Myrmonyssus* (*Laelaspulus*) Berlese, 1904 as a synonym of *Myrmozercon*. The only species known from western Asia and Eastern Europe are *M. ovatum* Karawajew, 1909 from Turkmenistan, *M. tauricus* Trach & Khaustov, 2011, from Ukraine and *M. Karajensis* Joharchi et al., 2011 from Iran. In this paper, we describe a new species of *Myrmozercon* found in Iran.

Materials and methods

Laelapidae associated with ants were collected mainly in Khorasan Razavi Province over a period of two years. Mites were removed from ants' nests by hand picking or by extraction from ant nesting material using Tullgren funnels. Mites were cleared in Nesbitt's solution and mounted in Hoyer's medium. The nomenclature used for the dorsal idiosomal chaetotaxy is that of Lindquist and Evans (1965), the leg chaetotaxy is that of Evans (1963a), the palp chaetotaxy is that of Evans (1963b), and names of other anatomical structures mostly follow Evans and Till (1979). We use the term "lyrifissures" to refer to slit-shaped sensilli, and "pore" for circular or oval-shaped cuticular openings of unspecified function. Holotype and paratypes of the new species are deposited in the Acarological collection, Department of Plant Protection, Yazd Branch, Islamic Azad University (YIAU); paratypes are also deposited in the Jalal Afshar Zoological Museum, College of Agriculture, University of Tehran, Iran (JAZM) and in the Australian National Insect Collection, CSIRO Ecosystem Sciences, Canberra, Australia (ANIC). All measurements in the descriptions are given in micrometres (µm).

Genus Myrmozercon Berlese

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

- *Myrmozercon* Berlese, 1902: 699. Type species *Myrmozercon brevipes* Berlese, 1902, by monotypy.
- *Myrmonyssus* Berlese, 1903: 16. Type species *Myrmonyssus diplogenius* Berlese, 1903, designated by Berlese, 1904 (synonymy by Rosario and Hunter 1988).
- *Myrmonyssus (Laelaspulus)* Berlese, 1904: 437. Type species *Myrmozercon acuminatus* Berlese, 1903, by original designation (synonymy by Shaw and Seeman 2009).
- *Parabisternalis* Ueckermann & Loots, 1995: 35. Type species *Parabisternalis yemeni* Ueckermann & Loots, 1995, by original designation (synonymy by Shaw and Seeman 2009).

Notes on the genus. The diagnosis of *Myrmozercon* used here is based on that of Shaw and Seeman (2009). Most species of Myrmozercon, including the type species M. brevipes, show moderate to strong hypertrichy on the dorsal shield. However, M. burwelli Shaw & Seeman, 2009 (24-25 pairs), and the new species, have a reduced dorsal chaetotaxy. All species appear to have asymmetrical and unpaired setae on the dorsal shield, which makes it difficult to recognise their homology except the new species. In most species the dorsal shield is reduced or truncated posteriorly to expose a strip of unsclerotised opisthonotal skin, but this is not true for every species. Species of Myrmozercon also vary in the presence or absence of metasternal setae st4, the sternal shield of new species is extended to which the sternal shield is fused with the endopodal plates, with three pairs of setae and three pairs of lyrifissures and metasternal setae (st4) absent. The leg chaetotaxy of Myrmozercon species is variable, and does not provide diagnostic characters that define the genus (Shaw and Seeman 2009) and this is very characteristic and fixed in the new species. The new species has one ventral seta on the palp trochanter the same as in most species of Myrmozercon. Shaw and Seeman (2009) described a swelling on the dorso-distal edge of the palp trochanter in several species, but this structure is not present in new species. This instability in morphology, and the edentate chelicerae and short peritremes of Myrmozercon, suggest that Myrmozercon is parasitic on its ant hosts, and not simply a commensal in its host's nests, but this has not been established experimentally. The specimens of new species were found clinging to the abdomen and head of the ants.

Results

Myrmozercon cyrusi Ghafarian & Joharchi, sp. n. urn:lsid:zoobank.org:act:5DD70D4C-1312-49FA-A881-C663EB04E98E http://species-id.net/wiki/Myrmozercon_cyrusi Figures 1–13

Type material. Holotype, female, Kenevist Rural District in the Central District of Mashhad County, Khorasan Razavi Province, Iran, 36.97' N, 59.68' E, alt. 945 m, 25 April 2012, A. Ghafarian coll., in nest of *Monomorium* sp. (in YIAU). Paratypes, four females, same data as holotype (in JAZM and ANIC).

Description of the female. Figures 1–13. *Dorsal idiosoma* (Fig. 1). Length 522–534. Dorsal shield length 488–500, width 420–436 (n = 5). Shield posteriorly truncate, not covering entire idiosoma, leaving a curved strip of unprotected skin posterior to setae J5, shield without distinct reticulate ornamentation over whole surface; with 33 pairs of setae, 21 podonotal (z2 absent), 12 opisthonotal (Z4, S5 absent) and Z5 in soft skin posterior to shield, almost all setae except j1 and J4 slightly barbed in apical third or less, with club-like tip (Fig. 2), opisthonotal setae very long, reaching well past base of next posterior setae, dorsal shield setae increasing in length from anterior to posterior (j1 25-27, J1 54-59, J2 67-69, J3 79-82), without unpaired and asymmetrical seta, setae on shield uniform in length and thickness except j1 (25-27) and J4 (20-25)



Figures 1–13. *Myrmozercon cyrusi* Ghafarian and Joharchi sp. n., female. 1 Dorsal shield 2 Dorsal seta enlarged (J5) (not to scale) 3 Ventral idiosoma 4–5 Opisthogastric setae enlarged (not to scale) 6 Hypostome 7 Epistome 8 Chelicera 9 femur, genu and tibia I, dorsal aspect 10 femur, genu and tibia II, dorsal aspect 11 femur, genu and tibia III, dorsal aspect 12 femur, genu and tibia IV, dorsal aspect 13 Insemination structures.

very fine and minute. A pair of very fine and minute setae in R series on the lateral soft skin but appear on ventral view. Shield with eight pairs of minute pores and lyrifissures including a pair of lyrifissures situated near z1, other pores inconspicuous.

Ventral idiosoma (Fig. 3). Tritosternum with short broad base (10-11 × 15-17 wide) fused to sternal shield, bifurcated at a short distance above suture, laciniae 37-40 in length, with smooth edges, strap-like and broad at base; pre-sternal shields fused with sternal shield. Sternal shield (length 248-255) narrowest between coxae II (104-108) widest between coxae II and III (218-22), with biconvex anterior margin and extending beyond level of st1, lateral margins thickened and posterolateral corners fused with endopodal shield; posterior margin concave; shield bearing three pairs of smooth pointed setae (st1 30-37, st2 40-45, st3 50-51) and two pairs of lyrifissures, one pair between setae st1 and st2 and the other between st2 and st3; surface with indistinct reticulate ornamentation. Seta st4 absent, metasternal pores also on extent of sternal shield but metasternal plates apparently absent. Genito-ventral shield wide, strongly tapering posteriorly, 320-346 long, 168-174 maximum width. Surface of shield smooth with longitudinal markings in anterior half; with one pair of simple setae st5 (35-37). Anal shield triangular, its anterior without lineate ornamentation, cribrum small, anal pores indistinct, bearing short post-anal seta 15-17 long, and a pair of para-anal setae 37-42 long. Opisthogastric skin with long, narrow metapodal plates $(40-44 \times 8-10 \text{ wide})$ and eight pairs of setae, almost all setae slightly barbed in apical third or less, each arising on small sclerotised platelet (Figs. 4,5), (Jv1 47-50, Iv2 35-37, Jv5 64-73, Zv1 37-42, Zv2 45-50, Zv3 55-63, Zv4 45-50, Zv5 67-75). Peritreme very short (35-40), extending to posterior level of coxae III. Peritrematal shields absent, post-stigmatal section conspicuous, with one pair of pore.

Gnathosoma. Hypostomal groove with nine rows of denticles, 10 to 15 very fine denticles per row (Fig. 6). Hypostome with three pairs of setae, internal posterior hypostomal setae h3 longest, palp coxal setae absent; surface of hypostome ornamented with transverse and curved lines. Palp chaetotaxy: trochanter 1, femur 5, genu 5, tibia 12; all palp setae pointed, palp tarsal claw two-tined, dorsodistal edge of palp femur without swelling. Epistome triangular, smooth, with pointed apex (Fig. 7). Chelicera hyaline, fixed digit of chelicera reduced, with four minute terminal denticles, pilus dentilis, dorsal lyrifissure present (Fig. 8); movable digit weakly sclerotised, distally curved, with one small subterminal tooth and one stronger terminal tooth, cheliceral seta absent, arthrodial corona with hyaline flap without filaments (Fig. 8). Corniculi long, weakly sclerotised.

Legs: Legs II and III short (258–268, 268–272), I and IV longer (288-298). Chaetotaxy: Leg I: coxa 0 0/1 0/1 0, trochanter 1 0/1 1/1 1 (*pd* thick), femur 1 2/1 2/1 1 (*ad*1 and *pd*1 long, ventral setae all thick, Fig. 9), genu 1 2/1 2/1 1 (*ad*1 and *pd*1 long with club-like tip, ventral setae all thick, Fig. 9). Leg II: coxa 0 0/1 0/1 0, trochanter 1 0/1 0/2 1, femur 1 2/1 2/1 1 (*ad*1 and *pd*1 thick, Fig. 9). Leg II: coxa 0 0/1 0/1 0, trochanter 1 0/1 0/2 1, femur 1 2/1 2/1 1 (*ad*1 and *pd*1 thick, *al* and *pl* long, Fig. 10), genu 1 2/1 2/1 1 (*ad*1 thick, *pd*1 long with club-like tip, *al* and *pl* long, Fig. 10), tibia 1 1/1 2/1 1 (*al* and *pl* long, Fig. 10). Leg III: coxa 0 0/1 0/1 0, trochanter 1 0/1 0/1 1 (*al* thick), *femur* 1 2/1 1/1 1 (*ad*1 thick, *al* and *pl* long, Fig. 11), genu 1 2/1 2/1 1 (*ad*1 and *pd*1 thick, *al* and *pl* long, Fig. 11), tibia 1 1/1 2/1 1 (*al* and *pl* long, Fig. 11). Leg IV: coxa 0 0/1 0/0 0, trochanter 1 1/2 0/1 0 (*ad* thick),

femur 1 2/1 1/1 0 (*al* long Fig. 12), genu 1 2/1 2/1 1 (*ad*1 and *pd*1 thick, *ad*1 in two paratypes club-like tip, *al* and *pl* long, Fig. 12), tibia 1 1/1 2/1 1 (*al* and *pl* long, Fig. 12). Tarsi I-IV with 16 setae, pre-tarsi with membranous ambulacrum, claws absent.

Genital structures: Insemination ducts opening on posterior margin of coxa III; sacculus an irregular, dark coloured mass behind coxae IV, ducts entering sacculus via a pair of circular openings (Fig. 13).

Etymology. The species is named in memory of Cyrus the Great (Old Persian: *Kuruš*; c. 600 BC or 576 BC–530 BC) was the first Achaemenian Emperor of Persia, as the "father of the Iranian nation", who issued a decree on his aims and policies, later hailed as his charter of the rights of nations.

Notes. *Myrmozercon cyrusi* differs from all other species in the genus by its very short peritreme, palp coxal setae absent, genua I-IV with similar chaetotaxy (1 2/1 2/1 1) and trochanter of palp with only one ventral seta.

Discussion

Only eleven species of *Myrmozercon* have been described from the Palaearctic Region (M. acuminatus (Berlese, 1903) on Messor capitatus (Latereille, 1798) from Italy; M. antennophoroides (Berlese, 1904) on Camponotus aethiops (Latereille, 1798) from Italy; M. brachiatus (Berlese, 1903) on Messor capitatus from Italy; M. brevipes Berlese, 1902 on Tapinoma erraticum (Latereille, 1798) from Italy; M. clarus (Hunter and Hunter, 1963) on Crematogaster clara Mayr from Georgia; M. diplogenius (Berlese, 1903) on Camponotus aethiops from Italy; M. flexuosa (Michael, 1891) on Camponotus herculeanus (L., 1758); M. Karajensis Joharchi et al., 2011 on Camponotus sp. from Iran; M. liguricus Vitzthum, 1930 on Crematogaster scutellaris (Olivier, 1792) from Germany; M. ovatum Karawajew, 1909 one from a worker Myrmecocystus emeryi Karawajew, 1909, but mostly on workers of Tapinoma erraticum nigerrimum from Turkmenistan; M. tauricus Trach & Khaustov, 2011 on Crematogaster schmidti (Mayr, 1853) from Ukraine). Three subfamilies and seven genera of ants have been reported as hosts from the world: Formicinae, Camponotus, Cataglyphis, Polyrhachis; Dolochoderine, Iridomyrmex, Tapinoma; Myrmecinae, Crematogaster, Messor. Myrmozercon cyrusi has been collected in association with *Monomorium* sp. and this is the first record of ant host.

According to publications, *M. ovatum* Karawajew, 1909 shares many compelling characters with *M. brevipes* Berlese, 1902 especially form of genital shield, short peritreme, short legs, dorsal shield highly hypertrichous and collecting on same host, but we have not had the opportunity to examine type specimens of these two species therefore we consider the *M. ovatum* to be a suspected synonym of *M. brevipes*.

The biology of *Myrmozercon* species has not been studied yet. However, instability in morphology, the edentate chelicerae and short peritremes might suggest that *Myrmozeron* is parasitic on its ant hosts, and not simply a commensal in its host's nests, but this has not been established experimentally.

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RESEARCH ARTICLE



Sinularia leptoclados (Ehrenberg, 1834) (Cnidaria, Octocorallia) re-examined

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Abstract

Sinularia leptoclados (Ehrenberg, 1834) is re-described. *Sinularia leptoclados* var. *gonatodes* Kolonko, 1926 is synonymized with *S. maxima* Verseveldt, 1977. Two new species of *Sinularia* with digitiform lobules, *leptoclados*-type surface clubs and unbranched interior spindles, are described. An updated maximum like-lihood tree of *Sinularia* species with *leptoclados*-type clubs (clade 5C) based on two mitochondrial genes (*mtMutS, COI*) and a nuclear gene (28S rDNA) is presented.

Keywords

Alcyonacea, re-description, new species, Indo-Pacific, Red Sea, taxonomy, phylogeny

Introduction

In his revision of the soft coral genus *Sinularia*, Verseveldt (1980) mentioned three stalked *Sinularia* species with digitiform lobules, *leptoclados*-type surface clubs and unbranched interior spindles. These are *Sinularia firma* Tixier-Durivault, 1970, *S. lepto-clados* (Ehrenberg, 1834), and *S. maxima* Verseveldt, 1971. Subsequently, comparison

of material collected from New Caledonia (RMNH Coel. 10447-10449) to type material proved *S. firma* to be an encrusting species. The original description of *S. firma* was based on a tiny fragment that obscured the colony growth form. In the key of Verseveldt (1980: 12) *S. leptoclados* and *S. maxima* were separated from each other by colony growth form, *S. maxima* with robust lobes, up to 120 mm high, and *S. leptoclados* with shorter ones. Verseveldt based his description of *Sinularia leptoclados* on a RMNH specimen from the Red Sea, without mentioning its catalogue number. He certainly did not have the type specimen, as he states that he failed to find that in museum collections (Verseveldt: 9). Additionally, in his revision Verseveldt synonymized *Sinularia leptoclados* var. *gonatodes* Kolonko, 1926, with *S. leptoclados* and stated the species exhibited an Indo-Pacific distribution.

The first two authors have based their identifications of *S. leptoclados* on the microscope slides of Verseveldt at their disposal, and following the *Sinularia* revision of Verseveldt (1980), have considered *S. leptoclados* specimens to be stalked with finger-like lobules and variable *leptoclados*-type clubs in the surface layer of the colony. The results have been published in a series of studies (see below) that have further supported Verseveldt's (1980) statement that the species is widespread in the Red Sea and in the Indo-West Pacific area.

Alderslade and Shirwaiker (1991) were the first after Verseveldt's (1980) revision to describe another species with characters similar to *S. leptoclados*, their *S. kavarattiensis* from the Laccadive Archipelago, India. They compared *S. kavarattiensis* with the holotype of *S. leptoclados* var. *gonatodes* and considered the many small spindles present in the surface layer of the lobes of the latter as a major difference between the two species. Later on, Manuputty and Ofwegen (2007) described three species from Ambon (Indonesia) which resembled *S. leptoclados*: *S. acuta, S. corpulentissima* and *S. longula.* In that study they used for comparison a specimen from Ambon (RMNH Coel. 38426), considered by them to be *S. leptoclados*.

McFadden et al. (2009), the first molecular study of the genus Sinularia ever conducted, discovered that specimens from Australia identified as S. leptoclados by P. Alderslade (NTM C5421) and the first author (NTM C14492, 14519-21) differed genetically from Red Sea specimens identified as S. leptoclados by the second author (ZMTAU CO 34095). This unexpected finding prompted us to re-examine the S. leptoclados collections of the RMNH and ZMTAU and to search for the type material of this widespread species (e.g., Verseveldt). Fortunately, we discovered the type specimen of S. leptoclados still exists in the ZMB, probably overlooked by Verseveldt, while revising the genus, most likely because it was labelled as Lobularia leptoclados Ehrenberg, 1834. After examination of its sclerites and comparison to RMNH and ZMTAU material identified as S. leptoclados it became obvious that this species does not exhibit an Indo-West Pacific distribution as stated by Verseveldt (1980), but is rather limited to the Red Sea and eastern Indian Ocean. Material wrongly assigned to S. leptoclados from other parts of the Indo-Pacific by the two first authors proved to be a mixture of misidentifications and as yet undescribed species. Interestingly, the specimens from Australia that were erroneously identified as S. leptoclados have sclerites and a colony morphology that closely resemble that

species. However, certain small morphological differences, its unique genetic haplotype, and the now disjunct distribution (Red Sea and western Indian Ocean *vs.* Pacific Ocean, Australia), convinced us to describe this material as a new species.

While collecting new material of *S. leptoclados* at Eilat, northern Gulf of Aqaba, Red Sea, we unexpectedly found two other species with *leptoclados*-type clubs and *leptoclados*-like colony shape: *S. verseveldti* Ofwegen, 1996 (Fig. 5f), so far only known from the Pacific, and a yet undescribed species which is described here.

Material and methods

Morphological examination

In order to identify the material, sclerites from different parts of the colony were obtained by dissolving the tissues in 10% sodium hypochlorite, followed by rinsing in fresh water. When appropriate, they were prepared for scanning electron microscopy as follows: the sclerites were carefully rinsed with double-distilled water, dried at room temperature, coated with gold and examined with a Jeol 6480LV electron microscope, operated at 10 kV.

Material studied is deposited in the Naturalis Biodiversity Center (formerly Rijksmuseum van Natuurlijke Historie, Leiden, the Netherlands (RMNH)), Zoological Museum, Department of Zoology, Tel Aviv University, Israel (ZMTAU), Museum für Naturkunde der Humboldt-Universität, Berlin, Germany (ZMB), Zoological Reference Collection (ZRC) of the Raffles Museum of Biodiversity Research, Singapore, and the Museum and Art Gallery of the Northern Territory, Darwin, Australia (NTM).

Molecular phylogenetic analysis

Extraction of DNA from ethanol-preserved tissue samples, PCR amplification, and sequencing of the *mtMutS* (*msh1*), *COI* and 28S rDNA genes followed the protocols published in McFadden et al. (2011) and McFadden and Ofwegen (2012). Sequence data were proofread using LaserGene software, and aligned using the L-INS-i method in MAFFT (Katoh et al. 2005). Pairwise measures of genetic distance (uncorrected p) among sequences were computed using MEGA v.5 (Tamura et al. 2011). Modeltest 3.0 (Posada and Crandall 1998) was used to select appropriate models of evolution for maximum likelihood analyses that were run using GARLI 2.0 (Zwickl 2006). Trees for *mtMutS* and *COI* were generally congruent with those for 28S rDNA, so in addition to separate analyses of the mitochondrial and nuclear genes we also ran a combined analysis with different models of evolution applied to each data partition (*mtMutS* + *COI*: TrN+I; 28S: GTR+I+G). Bayesian analyses of the same separate and combined data sets were run using MrBayes v. 3.2.1 (Ronquist et al. 2012) and a GTR+I+G model of evolution applied to both partitions; analyses were run for 2 million genera-

tions (until standard deviation of split partitions < 0.01) with a burn-in of 25% and default Metropolis coupling parameters. We included in our analyses all other species from *Sinularia* clade 5C for which sequence data were available for at least two of the three genes (Table 1); three species belonging to clades 5A (club sclerites with a distinct central wart, polyps with collaret, points and tentacle scales; *S. gardineri*) and 5B (club sclerites with a distinct central wart, polyps with collaret, polyps with collaret, points and tentacle scales; *S. hirta, S. terspilli*) were used as outgroup taxa.

Taxonomy

Sinularia australiensis sp. n.

urn:lsid:zoobank.org:act:C0EC77D7-A9DF-49A6-8BC4-C93AC3AFE8AF http://species-id.net/wiki/Sinularia_australiensis Figs 1–4

- ? Sinularia leptoclados; Lüttschwager, 1915: 3 (West Australia); Macfadyen: 37 (Great Barrier Reef Australia).
- *Sinularia leptoclados*; Ofwegen, 2008a: 131; McFadden et al.: 320 (Gulf of Carpentaria, West Australia).

Material examined. Holotype: NTM C14519, Australia, Northern Territory, Gulf of Carpentaria, West of Bremer island, 12°05.660'S, 136°47.754'E, depth 1–3 m, coll. P. Alderslade & party, 17 December 2003. Paratypes: NTM C14492, C14520, C14521, same data as holotype.

Description. The holotype is 6 cm high and 9.5 cm wide, attached to a piece of rock (Fig. 1A). The middle part of the colony is devoid of lobes, possibly a colony in the process of colony fission. The primary lobes branch off once or twice, lobules knobto finger-shaped, up to 4 mm wide and 1 cm long.

The polyps have a collaret and eight points. Points with poorly developed clubs, up to 0.15 mm long (Fig. 2A). Collaret has bent spindles, up to 0.20 mm long (Fig. 2B). Tentacle sclerites were not present.

The surface layer of the lobules has *leptoclados*-type clubs, the smallest are 0.07 mm long, most are around 0.10 mm, but some even reach a length of 0.15 mm (Fig. 2C); in addition, longer wart clubs are present, up to 0.25 mm long (Fig. 2D). Furthermore, the surface layer of the lobules has spindles, up to 0.40 mm long, with simple tubercles (Fig. 2E).

The sclerites of the surface layer of the base of the colony resemble those of the surface layer of the lobules but the clubs have wider handles and the spindles are wider (Fig. 3).

The interior of the colony has mostly unbranched spindles; a few have one or two side branches. In the lobules the spindles are up to 2.5 mm long (Fig. 4A), almost all having simple tubercles (Fig. 4B). In the base of the colony they are up to 3 mm long (Fig. 4C), with more complex tubercles (Fig. 4D).



Figure I. *Sinularia australiensis* sp. n., **A** holotype NTM C14519 **B** paratype NTM C14492 **C** paratype NTM C14520 **D** paratype NTM C14521. Scale at **A** also applies to **B**, scale at **C** also to **D**.

Colour. The preserved specimen is brown.

Etymology. Named after Australia, where the type was collected.

Intraspecific variation. NTM C14492 (Fig. 1B) and NTM C14521 (Fig. 1D) have stouter lobules, up to 1 cm wide.

Remarks. The species resembles *Sinularia leptoclados* regarding clubs and colony shape. It differs in having small surface lobule spindles with uniformly placed tubercles and many internal lobule spindles with simple tubercles. Other species resembling *S. australiensis* are *S. acuta* Manuputty & Ofwegen, 2007, S. *corpulentissima* Manuputty & Ofwegen, 2007 and *S. longula* Manuputty & Ofwegen, 2007, all three described from Ambon. *S. acuta* and *S. longula* have more slender spindles and wart clubs in the surface layer of the lobules (Manuputty and Ofwegen 2007: Figs 3, 19). *S. corpulentis*-



Figure 2. *Sinularia australiensis* sp. n., holotype NTM C14519. **A** point clubs **B** collaret spindles **C** *leptoclados*-type clubs of surface layer of lobule **D** wart clubs of surface layer of lobule **E** spindles of surface layer of lobule. Scale of 0.10 mm at **E** only applies to **E**.



Figure 3. *Sinularia australiensis* sp. n., holotype NTM C14519. Sclerites of the surface layer of the base of the colony **A** *leptoclados*-type clubs **B** wart clubs **C–D** spindles. Scale of 0.10 mm at **D** only applies to **D**.



Figure 4. *Sinularia australiensis* sp. n., holotype NTM C14519. sclerites of the interior **A** spindles from the lobules **B** tuberculation of one of the lobule spindles **C** spindles from the base **D** tuberculation of one of the base spindles. Scale of 1 mm at **C** also applies to **A**.
sima, like *S. leptoclados*, differs in having many internal spindles with complex tubercles (Manuputty and Ofwegen: Fig. 7c). Moreover, in the current molecular study *S. corpulentissima* is assigned to a distinct subclade together with *S. maxima*, while *S. acuta* and *S. longula* fall into a separate well-supported subclade (Figs 16, 17). *S. australiensis* sp. n. does not belong to either of those subclades, but is close genetically to *S. leptoclados* and *S. abrupta*. The latter species has clubs resembling those of *S. leptoclados* and *S. australiensis*, but a totally different colony shape, with ridges instead of lobes with lobules.

Lüttschwager (1915) and Macfadyen (1936) had *Sinularia* material from Australia that could belong to *S. australiensis*, but re-examination of sclerites of these specimens is necessary to confirm this possibility

Sinularia eilatensis sp. n.

urn:lsid:zoobank.org:act:2DE6BD04-F415-48CB-AFB5-ABF3EF9BA63D http://species-id.net/wiki/Sinularia_eilatensis Figs 5A–E, 6–9

Type material examined. holotype ZMTAU Co 35260, Israel, Red Sea, northern Gulf of Aqaba, Eilat, IUI (the Interuniversity Institute for Marine Sciences in Eilat) reef, depth 6 m, coll. Y. Benayahu, 10 January 2011; paratypes: ZMTAU Co 35261, same data as holotype; ZMTAU Co 35305, same data as holotype, 30 May 2011.

Other material examined: ZMTAU Co 35303-04, Israel, Red Sea, northern Gulf of Aqaba, Eilat, IUI reef, depth 5 m, coll. Y. Benayahu, 30 May 2011.

Description. The holotype is 3.4 cm high and wide (Fig. 5A). The primary lobes branch off once or twice, lobules finger-shaped, up to 2 mm wide and 1 cm long.

The polyps have a collaret and eight points. Points with poorly developed clubs, up to 0.25 mm long (Fig. 6A), collaret with bent spindles, up to 0.25 mm long (Fig. 6B) Tentacles with rods, about 0.05 mm long (Fig. 6C).

The surface layer of the lobules has *leptoclados*-type clubs, the smallest are 0.07 mm long, most are around 0.10 mm, but some reach a length of 0.15 mm (Fig. 6D); in addition longer wart clubs are present, up to 0.25 mm long (Fig. 6E). Furthermore, the surface layer of the lobules has spindles, up to 0.35 mm long, with simple tubercles (Fig. 6F).

The sclerites of the surface layer of the base of the colony resemble those of the surface layer of the lobules but they are wider (Fig. 7).

The interior of the colony has mostly unbranched spindles, a few have one or two side branches. In the lobules they are up to 2.5 mm long (Fig. 8A), with simple or complex tubercles (Fig. 8B). In the base of the colony the spindles are up to 2 mm long (Fig. 8C–D), with more complex tubercles (Fig. 8E).

Colour. The preserved holotype is dark brown.

Etymology. Named after Eilat, the type locality.



Figure 5. *Sinularia eilatensis* sp. n., colonies. **A** ZMTAU Co 35260, holotype **B** ZMTAU Co 35261, paratype **C** ZMTAU Co 35305, paratype **D** ZMTAU Co 35303 **E** ZMTAU Co 35304 **F** *S. verseveldti*, ZMTAU Co 35309.

Intraspecific variation. ZMTAU Co 35305 (Fig. 5C) has distinctly longer lobules, up to 2 cm long.

Remarks. The species is unique among *Sinularia* species with *leptoclados*-type clubs by its very long point and collaret sclerites.

We excluded ZMTAU Co 35303-04 (Fig. 5D–E) from the type series. Morphologically we could not find a difference between these two specimens and the types, but their mitochondrial gene haplotypes differ by 0.5%. For comparison, we also present sclerites of ZMTAU Co 35304 (Fig. 9).



Figure 6. *Sinularia eilatensis* sp. n., holotype ZMTAU Co 35260. **A** point clubs **B** collaret spindles **C** tentacle rods **D** *leptoclados*-type clubs of surface layer of lobule **E** wart clubs of surface layer of lobule **F** spindles of surface layer of lobule. Scale of 0.10 mm at **F** only applies to **F**.



Figure 7. *Sinularia eilatensis* sp. n., holotype ZMTAU Co 35260. Sclerites of the surface layer of the base of the colony. **A** *leptoclados*-type clubs **B** wart clubs **C** spindles.



Figure 8. *Sinularia eilatensis* sp. n., holotype ZMTAU Co 35260. Sclerites of the interior **A** spindles from the lobules **B** tuberculation of one of the lobule spindles **C–D** spindles from the base **E** tuberculation of one of the base spindles. Scale at **D** only applies to **D**.



Figure 9. *Sinularia eilatensis* sp. n., ZMTAU Co 35304. **A** point clubs **B** collaret spindles **C** *leptoclados*type clubs of surface layer of lobule **D** wart clubs of surface layer of lobule **E** spindles of surface layer of lobule **F** interior spindles of lobule **G** tuberculation of one of the lobule spindles. Scale of 0.10 mm at **E** only applies to **E**, 1 mm scale at **F** only to **F**.

Sinularia leptoclados (Ehrenberg, 1834)

http://species-id.net/wiki/Sinularia_leptoclados Figs 10A–E, 11–14

Lobularia leptoclados Ehrenberg, 1834: 58 (Red Sea).

- Alcyonium leptoclados; Klunzinger 1877: 26, pl. 1 fig. 7a-d (Red Sea).
- Sinularia leptoclados; Tixier-Durivault 1951: 124, figs 173-175 (Red Sea); 1966: 218, 222, figs 212–214 (Madagascar); Verseveldt 1965: 29 (Red Sea); 1971: 4 (Madagascar); Ofwegen and Benayahu 1992: 140 (Tanzania); Benayahu and Schleyer 1996: 6 (Mozambique); Benayahu et al. 2002: 278 (Southern Red Sea).
- NOT *Alcyonium leptoclados*; Burchardt 1903: 661, pl. 54 fig. 6, pl. 56 fig. 4 (Torres Strait, Ambon).
- NOT Sinularia leptoclados; Thomson and Dean 1931: 45, pl. 11 fig. 5, pl. 21 figs 6, 9 (Indonesia); Roxas 1933: 350, pl. 2 fig. 8 (Philippines); Verseveldt 1974: 96 (New Caledonia); 1977: 3 (Gambier Island, Fanning Atoll, Enewetak); 1978: 50 (Guam); Ofwegen and Vennam 1994: 138 (Ambon, Indonesia); Benayahu 1993: 6 (South Africa); 1995: 107 (Ryukyu Archipelago, Japan); Ofwegen 1996: 208 (Bismarck Sea); Benayahu 1997: 210 (Guam); Benayahu 2002: 14 (Ryukyu Archipelago, Japan); Benayahu et al. 2004: 551 (Taiwan); Manuputty and Ofwegen 2007: 192, figs 2b, 5 (Ambon, Indonesia; = S. verseveldti); Ofwegen 2008a: 131 (Gulf of Carpentaria, Australia; = S. australiensis sp. n.).
- NOT Sinularia aff. leptoclados Ofwegen, 2008b: 671 (Palau; = S. verseveldti).
- NOT *Sinularia leptoclados* var. *gonatodes* Kolonko, 1926: 309, pl. 2 fig. 1 (Philippines); Roxas 1933: 351 (same data as Kolonko) (= *S. maxima* Verseveldt, 1971)
- ? Sclerophytum herdmanni Pratt, 1905: 235, pl. 2 figs 8–9 (Sri Lanka; needs re-examination).

Material examined. ZMB 304, holotype of *Lobularia leptoclados* Ehrenberg; 1834, Rotes Meer, leg. Hemprich. Additional material: **Red Sea;** ZMTAU Co 25763, Egypt, Sinai, Tiran Strait, Thomas W., depth 3 m, coll. Y. Benayahu, 25 June 1985; ZMTAU Co 25940, Egypt, Gulf of Suez, Jubal Island, Bluf Point, depth 16 m, coll. Y. Benayahu, 24 March 1988; ZMTAU Co 34093-95, Israel, Gulf of Aqaba, Eilat, Nature Reserve, 29°30.6'N, 34°55.35'E, depth 2.4–5.5 m, coll. Y. Benayahu, 24 July 2007; ZMTAU Co 35308, Israel, Gulf of Aqaba, Eilat, Nature Reserve, depth 3 m, coll. Y. Benayahu, 31 May 2011; **Kenya;** ZMTAU Co 30354, off Mombasa, Shelly Reef, 04°07'S, 39°40'E, depth 12–13 m, coll. Y. Benayahu & S. Perkol, 20 January 2000; ZMTAU Co 32549, Shimoni, Wasini Is., opposite the building, depth 5 m, coll. Y. Benayahu, 2 February 2003; **Tanzania;** RMNH Coel. 18953, off Dar es Salaam, Pangavinne Island, seaward slope (P02), 6°50'S, 39°17'E, depth 6 m, coll. J.N. Nyanda; RMNH Coel. 18954, off Dar es Salaam, Pangavinne Island, seaward slope (P18), 6°50'S, 39°17'E, depth 8 m, coll. J.N. Nyanda; RMNH Coel. 18955, off Dar es Salaam, Mbudya Island, seaward slope (P35), 6°50'S, 39°17'E, depth 5 m, coll. J.N.



Figure 10. *Sinularia leptoclados* colonies. **A** ZMB 304 holotype **B** ZMTAU Co 34093 **C** ZMTAU Co 34094 **D** ZMTAU Co 34095 **E** ZMTAU Co 35308 **F** *Sinularia maxima*, ZRC1999.1066. Scale of 2 cm only applies to **A** and **F**.



Figure 11. *Sinularia leptoclados* holotype ZMB 304. **A** point clubs **B** *leptoclados*-type clubs of surface layer of lobule **C** wart clubs of surface layer of lobule **D** spindles of surface layer of lobule.



Figure 12. *Sinularia leptoclados* holotype ZMB 304. **A** spindles of the surface layer of lobule **B–D** sclerites of the interior **B** spindles from the lobules **C** tuberculation of one of the lobule spindles **D** spindles from the base **E** tuberculation of two of the base spindles. Scale of 0.10 mm at **A** only applies to **A**.



Figure 13. *Sinularia leptoclados* holotype ZMB 304. Sclerites of the surface layer of the base of the colony. **A** *leptoclados*-type clubs **B** wart clubs **C–D** spindles. Scale of 0.10 mm at **C** only applies to **C**.



Figure 14. Sinularia leptoclados colonies. A ZMTAU Co 25763 B ZMTAU Co 25940.

Nyanda; ZMTAU Co 26314, Pangavinne Is., depth 6 m, coll. J.N. Nyanda, 1991; ZMTAU Co 26316, Mbudya Is., depth 5 m, coll. J.N. Nyanda, 1991; **Mozambique;** ZMTAU Co 28796, Bazaruto Is., Manta Reef, depth 15 m, coll. M. Schleyer, 7 Oc-

tober 1994; **Madagascar**; RMNH Coel. 6653, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 22 July 1967, coll. A.G. Humes (1183); RMNH Coel. 6654, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 11 August 1967, coll. A.G. Humes (1250); RMNH Coel. 6655, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 23 August 1967, coll. A.G. Humes (1320); RMNH Coel. 6659, Nosy Iranja, SW Nosy Bé, depth 15 m, 9 August 1967, coll. A.G. Humes (1320); RMNH Coel. 6659, Nosy Iranja, SW Nosy Bé, depth 15 m, 9 August 1967, coll. A.G. Humes (1239); RMNH Coel. 6660, W of Andilana, 13°18'S, 48°07'E, 20 m deep, 24 August 1967, coll. A.G. Humes (1331); RMNH Coel. 6656, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 23 August 1967, coll. A.G. Humes (1321); RMNH Coel. 6657, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 23 August 1967, coll. A.G. Humes (1322); RMNH Coel. 6658, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 23 August 1967, coll. A.G. Humes (1322); RMNH Coel. 6658, Ankify, on mainland of Madagascar, opposite Nosy Komba, depth 1 m, 23 August 1967, coll. A.G. Humes (1323); RMNH Coel. 6661, Pass at Pte Lokobe, Nosy Bé, Madagascar, depth 15 m, 19 June 1967, coll. A.G. Humes (A28).

Description. The holotype is 18 cm high and 13 cm wide (Fig. 10A). The primary lobes branch off once or twice, lobules finger-shaped, up to 1 cm wide and 3 cm long.

The polyps have a collaret and eight points. Points with poorly developed clubs, up to 0.13 mm long (Fig. 11A), collaret with bent spindles. Tentacle sclerites not observed.

The surface layer of the lobules has *leptoclados*-type clubs, the smallest are 0.05 mm long, most are around 0.10 mm, but some reach a length of 0.15 mm (Fig. 11B); in addition longer wart clubs are present, up to 0.20 mm long (Fig. 11C). Furthermore, the surface layer of the lobules has spindles, up to 0.45 mm long, with simple tubercles (Fig. 11D, 12A); the smaller ones with a distinct median waist.

The interior of the colony has unbranched spindles. In the lobules they are up to 2.5 mm long (Fig. 12B), with simple or complex tubercles (Fig. 12C). In the base of the colony the spindles are also up to 2 mm long (Fig. 12D), with more complex tubercles (Fig. 12E).

The sclerites of the surface layer of the base of the colony resemble those of the surface layer of the lobules but they are wider (Fig. 13).

Colour. The holotype is brown.

Intraspecific variation. Most of the colonies of *S. leptoclados* are stalked and rarely feature an encrusting colony shape (Fig. 14).

Remarks. Verseveldt (1980) re-examined ZMB 6495, the type specimen of Kolonko's *S. leptoclados* var. *gonatodes* from the Bata islands (East coast of Palawan, Philippines), and considered it nothing else than *S. leptoclados* (Ehrenberg, 1834). Alderslade and Shirwaiker (1991) also re-examined ZMB 6495, assuming it was *S. leptoclados*, to differentiate their *S. kavarattiensis*. They noticed many small rods in the surface layer of the lobes. Unfortunately, neither Verseveldt nor Alderslade and Shirwaiker presented any figures of sclerites of *Sinularia leptoclados* var. *gonatodes*. Here we present such sclerites of the lobe surface (Fig. 15), which are more like those of *S. maxima* Verseveldt, 1971, and therefore we consider *S. leptoclados* var. *gonatodes* to be *S. maxima*. The main difference between *S. leptoclados* and *S. maxima* is not in the sclerites but concerns the much wider lobes of the latter (Fig. 10E). As a consequence,



Figure 15. *Sinularia leptoclados* var. *gonatodes* ZMB 6495 **A** point clubs **B** *leptoclados*-type clubs of surface layer of lobule **C** wart clubs of surface layer of lobule **D** spindles of surface layer of lobule.

Alderslade and Shirwaiker (1991) compared their S. *kavarattiensis* with S. *maxima* instead of with S. *leptoclados*. Their new species differs from both in having *leptoclados*-type clubs with an angle between the head and handle of about 90 degrees, thus considered to be valid.

One other species that can be confused with *S. leptoclados* is *S. verseveldti* Ofwegen, 1996. Its colony shape was described as being cup-shaped, but examination of many specimens from Indonesia showed that colony shape to be exceptional. Mostly the colonies resemble *S. leptoclados* very closely. Manuputty and Ofwegen (2007, fig. 2b, fig. 5) showed such a colony and its sclerites. The species differs in club shape, with the angle between the head and handle larger than 90 degrees in *S. leptoclados* and about 90 degrees in *S. verseveldti*. *S.* aff. *leptoclados* in Ofwegen (2009: 671) we now consider also to be *S. verseveldti*.

Molecular Results

Sequences for *mtMutS* and *COI* (including *igr1*) were available or newly obtained for 31 specimens representing 19 morphospecies of *Sinularia* belonging to clade 5C; 28S rDNA sequences were obtained for all but four specimens (Table 1). *mtMutS* (735 nt) and *COI* (888 nt) sequences were concatenated for a total mitochondrial gene alignment of 1623 nt. 28S sequences ranged from 797–799 nt in length for a total alignment length of 801 nt. Maximum likelihood and Bayesian analyses resulted in identical tree topologies for all three data sets (mt genes only, 28S only, all three genes combined). Support values were generally somewhat stronger for Bayesian analyses, however, and several nodes that were not supported by maximum likelihood (bootstrap values <50%) nonetheless had Bayesian posterior probabilities >0.9 (Figs 16, 17). All alignments and trees have been submitted to TreeBASE (www.treebase.org).

Within *Sinularia* species with *leptoclados*-type clubs (clade 5C), genetic distances (uncorrected p) among recognized morphospecies range from only 0-1.7% for *mt-MutS*, 0-0.8% for *COI* and 0-1.4% for 28S rDNA. Despite these relatively low levels of genetic differentiation among taxa, several moderately- to well-supported clades appear in both the mitochondrial and 28S gene trees (Fig. 16). *S. maxima* and *S. corpulentissima* share identical mt and 28S haplotypes with one another, but are well differentiated from all other species in clade 5C. *S. acuta, S. longula* and *S. molesta* are also very similar to one another genetically (*S. molesta* and *S. acuta* share identical mt and 28S haplotypes), and form a well-supported clade in both trees. Finally, *S. erecta* is genetically distinct, separated from all other species by genetic distances of >0.8% at *mtMutS* (28S was not available for *S. erecta*).

Two additional clades are moderately supported by the combined analysis of the mt and 28S genes (Fig. 17); the species in these clades also group together in the separate analyses, but with low bootstrap support (<50%) (Fig. 16). *S. penghuensis, S. bisulca, S. robusta, S. digitata* and *S. slieringsi* comprise one of these moderately-supported clades (Fig. 17); these species share identical or nearly identical 28S sequences

		GenBank Acc. No.		
Species	Museum Acc. No.	ICO	mtMutS	28S rDNA
S. abrupta	NTM C14012	KC542862	KC542849	NA
S. abrupta	ZMTAU Co 33623	JX991256	JX991168	KC542822
S. acuta	RMNH Coel. 38721	KC542863	FJ621376	NA
S. acuta	ZMTAU Co 33617	JX991257	JX991169	KC542823
S. australiensis sp. n.	NTM C14492	KC542864	FJ621437	KC542824
S. australiensis sp. n.	NTM C14519	KC542865	FJ621438	KC542825
S. bisulca	RMNH Coel. 38724	KC542866	FJ621378	KC542826
S. corpulentissima	RMNH Coel. 40839	KC542867	KC542850	KC542827
S. daii	ZMTAU Co 34665	JX991258	JX991170	KC542828
S. densa	RMNH Coel. 40840	KC542868	KC542851	KC542829
S. digitata	RMNH Coel. 40841	KC542869	KC542852	KC542830
S. eilatensis sp. n.	ZMTAU Co 35260	KC542870	KC542853	KC542831
S. eilatensis sp. n.	ZMTAU Co 35305	KC542873	KC542856	KC542834
S. ?eilatensis sp. n.	ZMTAU Co 35303	KC542871	KC542854	KC542832
S. ?eilatensis sp. n.	ZMTAU Co 35304	KC542872	KC542855	KC542833
S. erecta	ZMTAU Co 34144	GU355981	FJ621404	KC542835
S. gardineri (5A)	ZMTAU Co 34097	GU355982	FJ621414	KC542819
S. hirta (5B)	ZMTAU Co 34100	GU355983	FJ621428	KC542820
S. leptoclados	ZMTAU Co 35308	KC542874	KC542857	KC542836
S. leptoclados	ZMTAU Co 34095	GU355980	FJ621439	KC542837
S. longula	RMNH Coel. 38439	KC542875	FJ621441	KC542838
S. maxima	NTM C14512	KC542876	FJ621448	KC542839
S. molesta	RMNH Coel. 38440	KC542877	FJ621449	NA
S. penghuensis	ZMTAU Co 34659	JX991273	JX991183	KC542840
S. penghuensis	ZMTAU Co 34681	JX991274	JX991184	KC542841
S. penghuensis	ZMTAU Co 34739	JX991276	JX991186	KC542842
S. robusta	NTM C14518	KC542878	FJ621473	KC542843
S. slieringsi	ZMTAU Co 34654	JX991277	JX991187	NA
S. terspilli (5B)	ZMTAU Co 34156	GU355984	FJ621481	KC542821
S. verseveldti	ZMTAU Co 35309	KC542879	KC542858	KC542844
S. verseveldti	RMNH Coel. 40842	KC542880	KC542859	KC542845
S. verseveldti	RMNH Coel. 40843	KC542881	KC542860	KC542846
S. verseveldti	RMNH Coel.40844	KC542882	KC542861	KC542847
S. wanannensis	ZMTAU Co 34704	JX991281	JX991190	KC542848

Table 1. Specimens of *Sinularia* included in the molecular phylogenetic analyses. NTM = Museum and Art Gallery of the Northern Territory; RMNH = Naturalis Biodiversity Center; ZMTAU = Zoological Museum, Tel Aviv University. Bold = new GenBank accessions; NA = no sequence obtained.

(28S was not available for *S. slieringsi*) (Fig. 16b). Within the mt gene tree (Fig. 16a) they constitute two distinct clades, one comprised by *S. robusta, S. digitata* and *S. slieringsi* and the other by *S. penghuensis, S. bisulca* and *S. daii*. The latter is, however, distinct from all other species at 28S, and falls outside of this clade in the combined analysis (Fig. 17). *S. leptoclados, S. abrupta, S. australiensis* sp. n. and *S. densa* also

(a) mtMutS + COI

(b) 28S rDNA



Figure 16. Maximum likelihood trees of *Sinularia* clade 5C (McFadden et al. 2009) based on (**a**) combined analysis of two mitochondrial genes (*mtMutS*, *COI*), and (**b**) nuclear 28S rDNA. Specimens described in this publication in bold. Numbers above branches are bootstrap values from maximum likelihood analysis (only values >50% shown; ns = value <50%); numbers below branches are Bayesian posterior probabilities (only values > 0.85 shown).

form a moderately-supported clade in the 28S tree (Fig. 16b) and in the combined tree (supported by Bayesian but not maximum likelihood analyses; Fig. 17), but their relationship is unresolved in the mt tree (Fig. 16a). *S. australiensis* sp. n. and *S. abrupta* share identical 28S haplotypes, but differ from *S. leptoclados* by 0.3%. *S. australiensis* sp. n. differs from both *S. leptoclados* and *S. abrupta* by 0.1% and 0.1-0.2% at *mtMutS* and *COI* respectively.

The relationships among the remaining species in the clade — *S. verseveldti, S. wanannensis* and *S. eilatensis* sp. n. — were poorly resolved and exhibited some incongruence between the mitochondrial and 28S gene trees. *S. wanannensis*, all four specimens of *S. verseveldti*, and two specimens (ZMTAU Co 35303, ZMTAU Co 35304) that were tentatively assigned to *S. eilatensis* sp. n. share identical or nearly identical *mt*-



Figure 17. Maximum likelihood tree of *Sinularia* clade 5C (McFadden et al. 2009) based on a combined, partitioned analysis of two mitochondrial genes (*mtMutS, COI*) and nuclear 28S rDNA. Specimens described in this publication in bold. Specimens indicated with * have different mtDNA haplotypes but identical 28S rDNA sequences. Numbers above branches are bootstrap values from maximum likelihood analysis (only values >50% shown; ns = value <50%); numbers below branches are Bayesian posterior probabilities (only values > 0.85 shown).

MutS and *COI* haplotypes, and cluster together within the mt tree (but with bootstrap values <50%). Two specimens of *S. eilatensis* sp. n. (ZMTAU Co 35305, ZMTAU Co 35260) fall outside of that group, and differ from it by >0.5% at *mtMutS* (Fig. 16a). At 28S, however, ZMTAU Co 35303 and ZMTAU Co 35304 are genetically identical to both individuals of *S. eilatensis* sp. n., and those four specimens form a moderately-supported clade together with *S. verseveldti* ZMTAU Co 35309 (Fig. 16b). Two additional specimens of *S. verseveldti* share identical 28S haplotypes with *S. wanannensis*. The combined tree reflects the topology of the mt gene tree, and shows the separation of *S. eilatensis* sp. n. (ZMTAU Co 35305, ZMTAU Co 35200) from ZMTAU Co 35303, ZMTAU Co 35304 and all other species (Fig. 17).

Our findings indicate that specimens of the same species generally shared identical or nearly identical sequences at all three loci. The only exceptions were the two distinct mitochondrial haplotypes of *S. eilatensis* sp. n. discussed above, and the four specimens of *S. verseveldti*. All *S. verseveldti* shared identical or nearly identical *mtMutS* and *COI*

sequences, but differed at 28S. Most of these differences, however, reflected polymorphic nucleotide positions at which one or more specimens exhibited heterozygosity. For example, at position 533 of the 28S alignment, ZMTAU Co 35309 and Coel. 40842 had C, Coel. 40843 had T, and Coel. 40844 had both C and T. A total of 8 such heterozygous nucleotide sites among the four *S. verseveldti* specimens contribute to their disjunct distribution within the 28S and combined trees.

Discussion

The two new species described here are supported both by morphological characters and by the molecular analysis. Although *S. australiensis* sp. n. is similar genetically to *S. lepto-clados* and both belong to the same sub-clade within *Sinularia* clade 5C, they differ at all three of the loci sequenced here. Furthermore, the 28S and combined analyses suggest that *S. leptoclados* and *S. australiensis* sp. n. are not sister taxa, but that *S. australiensis* sp. n. is closer to *S. abrupta*, a species with which it shares a 28S haplotype. The disjunct geographical distribution between *S. leptoclados*, which occurs in the Red Sea and western Indian Ocean, and *S. australiensis* sp. n. from Australia, further supports their distinction.

Although sympatric with S. leptoclados in the Red Sea, S. eilatensis sp. n. is clearly distinct from that species, both morphologically and genetically. Within clade 5C, S. eilatensis sp. n. is most similar genetically to the geographically widespread S. verseveldti and to S. wanannensis, a species recently described from Taiwan (Ofwegen and Benayahu 2012). Morphologically, however, S. eilatensis clearly differs from S. verseveldti and S. wanannensis by its long polyp sclerites, up to 0.25 mm long in S. eilatensis vs up to 0.15 mm long in the other two species. Undoubtedly, the phylogenetic relationships among these three species need further investigation. In particular, the conflicting phylogenetic signals obtained from the mitochondrial and nuclear genes suggest the possibility of past hybridization events between S. verseveldti and S. eilatensis sp. n. Two specimens from the Red Sea (ZMTAU Co 35030 and ZMTAU Co 35304) appear morphologically to belong to S. eilatensis sp. n. and have the same 28S sequence as that species but share a distinct mitochondrial haplotype with S. verseveldti. This observed mito-nuclear discord could reflect a hybrid origin of these specimens, as has been suggested for some other octocorals (reviewed in McFadden et al. 2010). In addition, the polymorphism observed at the 28S locus in S. verseveldti could be indicative of recent hybridization events involving this species, although it could also be the result of incomplete lineage sorting following recent speciation. The possible hybrid origin of ZMTAU Co 35303 and ZMTAU Co 35304 should be investigated further using single-copy nuclear gene markers.

Previous molecular systematic work on *Sinularia* and other octocoral genera has highlighted the inadequacies of mitochondrial gene markers for species discrimination and species-level phylogenetic analyses in the group (McFadden et al. 2009, 2011). Although both *mtMutS* and *COI* effectively distinguish genera and distinct clades within genera, neither gene is variable enough to distinguish all congeneric

species pairs unequivocally. The region of the nuclear 28S rDNA gene we sequenced exhibits somewhat greater variability than *mtMutS* in some genera of the family Alcyoniidae (Benayahu et al. in press), but did not distinguish among all of the morphospecies of *Sinularia* examined in the current study. Despite the relatively small genetic distances separating morphospecies and the low resolution of the resulting phylogenies, we believe the analysis presented here adequately supports the distinctions of the new species that are the focus of this study. Development of additional, more variable molecular markers, will be necessary in order to fully resolve the relationships among morphospecies in *Sinularia* clade 5C and to address the possibility of hybridization among them.

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RESEARCH ARTICLE



Heptageniidae (Insecta, Ephemeroptera) of Thailand

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Abstract

Nine genera and twenty-two species of heptageniid mayflies from Thailand are defined in this present work as well as one suggested further subgenus, *Compsoneuria (Siamoneuria) kovaci* (species "incertae sedis") including some particular characters. Taxonomic remarks, diagnoses, line drawings of key characters, distribution, habitat and biological data, and a larval key to the genera and species are provided. The chorionic eggs of eight genera and eight species were observed and shown using a scanning electron microscope.

Keywords

Mayflies, Heptageniidae, Thailand, key

Introduction

Heptageniidae is a family of mayflies with around 509 described species and distributed mainly in the Holoarctic, Oriental, and Afrotropical regions (Barber-James et al. 2008). Wang and McCafferty (2004: Part I) "analyzed the generic relationships and presented a phylogenetic classification of the family" while Webb and McCafferty (2008: Part II) defined the genera providing an illustrated key. Following Webb and McCafferty (2008), only 16 genera but more than 150 species of Heptageniidae can be found in the Oriental region (Soldán, 2001).

Heptageniid mayflies are one of the most abundant and common components of benthic communities in Thai running waters. The larvae inhabit slow to fast flowing streams where they occur on the surface of rocks, logs, vegetation, and leaves. Many heptageniid species have been used as indicators of anthropogenic disturbance because they are relatively intolerant of pollution change and as sensitive indicators of organic pollution (Hilsenhoff 1988) and metal pollution (Courtney and Clements 1998, Deacon et al. 2001, Clements 2004, Clark and Clements 2006). Furthermore, individuals of this family were test subjects of toxicity and drift behavior studies (Diamond et al. 1992, Céréghino et al. 2004, Stitt et al. 2006).

Heptageniidae have been recorded from Thailand by Polhemus and Polhemus (1988), Braasch (1990), Sites et al. (2001), Sangpradub et al. (2002), Wang and McCafferty (2004), Braasch (2006a), Webb and McCafferty (2006), and recently by Braasch and Boonsoong (2009, 2010). However, taxonomic revision of the family Heptageniidae in Thailand is urgently needed, because the study of life stages is still in its infancy. This is mainly due to problems of identification, unsettled generic questions, and the lack of use of modern genetic methods to construct a phylogeny of the family Heptageniidae from Southeast Asia. In this paper, we provide a larval key to known genera and species of Thai heptageniid mayflies, with particular emphasis on the problems of identification of several species. Taxonomic remarks, diagnoses, line drawings of key characters, distribution, and habitat and biological data are provided. In addition, the egg chorions of eight Thai heptageniid species were observed. All of the egg specimens used in this study were obtained from mature larvae and adults. The material was first preserved in alcohol and then critical-point dried using carbon dioxide and finally placed on holders and coated with gold. The oological observations of eight heptageniid species were made with a JEOL JSM-5600LV scanning electron microscope. The terminology provided by Koss and Edmunds (1974) is used in this paper.

In the following key and text, abbreviations are as follows: alt (altitude), asl (above sea level), μ S/cm (microsiemens per centimeter), comb. (combination), M (male imago), F (female imago), mount. (mountainous), Ms (male subimago), Fs (female subimago), NP (National Park), orig. (original), sec. (second), WS (Wildlife Sanctuary).

A Larval Key to the Genera, Subgenera and Known Species of Heptageniidae in Thailand

1	Median caudal filament absent (Fig. 1A, 9D) Epeorus,	2
_	Median caudal filament present	8
2	Lamellae of gills 2-7 with anal ribs arched (Fig. 1B) subgenus Belovius,	4
-	Lamellae of gills 2-7 without anal ribs arched	3

3	Lamellae of gills 1 greatly extended beneath the abdomen (Fig. 1C–D)
	subgenus Iron, 5
_	Lamellae of gills 1 somewhat extended beneath the abdomen (Fig. 11)
/	subgenus <i>Epeorus</i> , 6
4	Abdominal terga $2-9$ each with long, acute median spine on posterior margin
	(Fig. 1F) E. unicornutus Braasch
_	Abdominal terga 2–9 each without acute median spine on posterior margin $(E_{1}, 1C)$
5	(Fig. IG) E. Rnayengensis Boonsoong & Braasch
)	<i>E mantinua</i> Brosseh & Soldán
	Abdominal terga without paired acute submedian spines: foretibiae relatively
_	long: median dark brown on the abdominal terga
	<i>F</i> thailandensis Braasch & Boonsoong
6	Pair of submedian spines on terga 2-9 relatively long (Fig. 1A)
0	<i>F. aculeatus</i> Braasch
_	Pair of submedian spines on terga 2-9 relatively short
7	Paired tubercles on terga rounded, blunt bristles denselv rowed
,	<i>E. bifurcatus</i> Braasch & Soldán
_	Paired tubercles on terga more rounded, pointed bristles (Fig.1H, 1J)
	<i>E. inthanonensis</i> Braasch & Boonsoong
8	Gill-pairs 1 meet or overlap ventrally to form a friction disc (Fig. 2A)
_	Gill-pairs 1 not meeting ventrally and not forming a ventral friction disc 9
9	Lamellae of gills 1 minute, lamellae of gills 2-7 long, narrow and sharply
	pointed (Fig. 2B-E); ventral surface of maxillae with setae in row (Fig. 2F);
	abdominal terga with fan-shaped robust setae (Fig. 2G)
	<i>Trichogenia maxillaris</i> Braasch & Soldán
_	Lamellae of gills 1 similar in shape and size to other gills; gill lamellae usu-
	ally not as above; ventral surface of maxillae with scattered setae (Fig. 2H);
	abdominal terga with fine setae only10
10	Abdominal terga with median dorsal ridge (Fig. 21–J); claws with denticles
_	Abdominal terga without median dorsal ridge
11	Supracoxal sclerites rounded or bluntly pointed (Fig. 3A)
-	Supracoxal sclerites sharply pointed (Fig. 3B)
12	Anterior margin of head capsule distinctly thickened (Fig. 3C); posterolateral
	spines of abdomen well developed (Fig. 3D) <i>Inderosphyrus sinuosus</i> Navas
-	Anterior margin of head capsule not thickened; dorsal view of abdomen; pos-
12	terolateral spines of addomen small (Fig. 51, K) Compsoneuria, 13
13	Shape of sills 3-6 with omercinations (E:~ 2E U)
_	Shape of glips 5-0 with emarginations (Fig. $5E-\Pi$)

14	Dorsal view of abdomen as Fig. 3I; shape of gill 7 leaf-like and pointed api-
	cally (Fig. 3J) C. thienemanni Ulmer
_	Dorsal view of abdomen as Fig. 3K; shape of gill 7 lanceolate and rounded
	apically (Fig. 3L) C. langensis Braasch & Boonsoong
15	Gills 7 slender and pointed (Fig. 4A); robust setae on inner surface of hind-
	tarsi pectinate (Fig. 4B)Asionurus primus Braasch & Soldán
_	Gills 7 usually rounded apically, never as long and narrow as above; setae on
	inner surface of tarsi either simple or fimbriate, never pectinate16
16	Cerci bear spines as well as lateral bristles and segments of the cerci with stout
	spines alternate with those lacking such spines (Fig. 4C); gills 1-7 with row
	of sparse marginal set Rhithrogeniella tonkinensis Soldán & Braasch
_	Cerci not as above (Fig. 4D); gills 1–7 without row of sparse marginal setae.
	Afronurus, 17
17	Body and head with indistinct large pale dots and markings dorsally18
_	Body and head with distinct large pale dots and markings dorsally (Fig. 4E) 19
10	
10	Gills 1 banana-shaped
18 -	Gills 1 banana-shaped
18 19	Gills 1 banana-shaped
18 19	Gills 1 banana-shaped

Taxonomic descriptions

Genus *Epeorus* Eaton, 1881

http://species-id.net/wiki/Epeorus Figs 1A–J, 5A–B, 9D

Remarks. *Epeorus* is widely distributed in the Holarctic, northern portion of the Neotropical, Palaearctic, and Oriental regions (Kluge 2004). In tropical Southeast Asia, species of this genus have been reported and described by Braasch (1990, 2011), Braasch and Boonsoong (2010), Braasch and Soldán (1979, 1984c), and Webb and McCafferty (2006). Nguyen and Bae (2004a) provided larval descriptions and a key to six species of Vietnamese *Epeorus* ; this was the first comprehensive taxonomic study of the larvae of *Epeorus* from tropical Southeast Asia. The first record of *Epeorus* in Thailand is of *E. aculeatus* Braasch (1990) from Doi Inthanon National Park, Chiang Mai province. Recently, the first Thai imago of *E. aculeatus* was described by Webb and McCafferty (2006) from Chiang Mai province. Originally, *E. unicornutus* was recorded from Himalayas (Braasch 2006b) although it had been already collected in Thailand December 1987/1998 from the river Nam Lang, Soppong / Pangmapa, Mae Hong Son province (Braasch, unpublished). Currently, seven species of *Epeorus* are known from Thailand (Braasch and Boonsoong 2010).

64



Figure 1. A Habitus of *Epeorus aculeatus* Braasch, 1990 **B** lamellae of gills 7 of *E. khayengensis* Boonsoong & Braasch, 2010 **C–E** ventral view of abdomen (**C**), abdominal gills 1 (**D**) and abdominal terga of (**E**) *E. thailandensis* sp. n. **F** abdominal terga of *E. unicornutus* Braasch, 2006 **G** abdominal terga of *E. khayengensis* Boonsoong & Braasch, 2010 **H–J** abdominal terga (**H**), lamellae of gills 1 (**I**) and tergum VII (**J**) of *E. inthanonensis* Braasch & Boonsoong, 2010.

Epeorus (Belovius) khayengensis Boonsoong & Braasch, 2010

http://species-id.net/wiki/Epeorus_khayengensis Figs 1B, 1G, 5A–B, 9D

Epeorus (Belovius) khayengensis Boonsoong & Braasch, 2010: 13–17, Figs 53–68. (orig.)

Larva. Braasch and Boonsoong 2010: 13-17, Figs 53–68. Adult. Unknown.

Eggs. Egg chorion of *E. khayengensis* very smooth without any peculiar structure (Fig. 5A), 2-3 micropyles visible in the equatorial area (Fig. 5B).

Distribution. Huai Khayeng stream (Thong Pha Phum district, Kanchanaburi province).

Diagnosis. The larva of *E. khayengensis* (Fig. 9D) can be distinguished from that of other congeners by abdominal terga 2–9 without long acute median spine on posterior margin, but bearing long hair-like setae.

Habitat and biology. The larva of *E. khayengensis* inhabits tropical headwater streams approximately 210 m in alt. The streams range between 6–7 m in width and 10–11 cm in depth. The water temperature ranges between 22–25°C, pH between 6.35–7.15, total dissolved solids are between 27–34 mg/L, and conductivity is between 41–53 μ S/cm. The larvae are found in eroded areas of streams where moderately flowing over cobble and sandy bottom.

Remarks. Braasch and Boonsoong (2010) described this species from Thailand based on nymphal specimens, and deposited them in the Zoological Museum, Kasetsart University (ZMKU), Bangkok. The adults of *E. khayengensis* are unknown.

Epeorus (Belovius) unicornutus Braasch, 2006

http://species-id.net/wiki/Epeorus_unicornutus Fig. 5F

Epeorus (Belovius) unicornutus Braasch, 2006: 80, 82, Figs 1-8. (orig.)

Larva. Braasch 2006b: 80, 82, Figs 1-8.

Adult. Unknown.

Eggs. Unknown.

Distribution. Nam Thob Ranger Station, Phu Luang Wildlife Sanctuary, Nam Thob stream (Loei province).

Diagnosis. The larva of *E. unicornutus* can be distinguished from that of other congeners by the combination of the following characters: abdominal terga 2-9 each with single, prominent, acute median spine and with a row of short spines on posterior margin, tergum 10 with short spines and hair-like setae on posterior margin.

Habitat and biology. Larvae of *E. unicornutus* are found in headwater streams shaded under tree canopies in mountainous areas (alt 330 m) where the streams are 10–12 m wide and 10–15 cm in depth. The water temperature ranges between 22–23°C, pH between 7.0–7.2, total dissolved solids range between 18–20 mg/L, and conductivity between 28–30 μ S/cm. The larvae are found underneath stones in fast flowing reaches of the streams. The coarse mineral substrate consists of boulder (60%), cobble (30%), gravel and coarse sand (10%), and abundant fallen leaves. *E. unicornutus* is found under minimally disturbed conditions of Nam Thob streams, Loei province.

Remarks. Braasch (2006b) described this species based on larval specimens from River Indravati near Dalaghat 1,200 m, Nepal, Himalayas. In this study, this species were found from northern and northeastern parts of Thailand. This species is different from other known species of the genus *Epeorus* by abdominal terga 2-9 each with single, prominent, and acute median spine.

Epeorus (Iron) martinus Braasch & Soldán, 1984

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

Iron martinus Braasch & Soldán, 1984: 113-114, Figs 35–44. (orig.) Epeorus (Iron) martinus Braasch & Soldán, 1984 (comb.)

Larva. Braasch and Soldán 1984c: 113–114, Figs 35–44; Nguyen and Bae 2004a: 102–104, Figs 1–6.

Adult. Unknown. Eggs. Unknown.

Distribution. Khun Kon Waterfall (Chiang Rai province).

Diagnosis. The larva of *E. martinus* can be distinguished by the following characteristics: pairs of moderately long acute submedian spines on the abdominal terga 1-9, large but narrow gill 1 forming a sucking disc and gill 7 being unfolded.

Habitat and biology. *E. martinus* larvae are found in mountain streams with a moderate current, at an elevation of 300–2800 m (Nguyen and Bae 2004a).

Remarks. Webb and McCafferty (2008) did not recognize the subgenera of *Epeorus* (e.g. *Belovius*, *Iron*). However, we hold on the validity of subgenus *Belovius* and *Iron* within genus *Epeorus* (Boonsoong and Braasch, 2010). Therefore, we propose *Iron martinus* Braasch & Soldán, 1984 = *Epeorus (Iron) martinus* Braasch & Soldán, 1984

Epeorus (Iron) thailandensis Braasch & Boonsoong, sp. n. urn:lsid:zoobank.org:act:1BCDC6C6-F254-4C28-ABBB-CF520AA3A2E4 http://species-id.net/wiki/Epeorus_thailandensis Figs 1C-E

Iron longitibius Nguyen & Bae, 2004 (questionable): 20–22, Figs 83–98. *Epeorus (Iron) thailandensis* Braasch & Booonsoong sp. n.

Larva. Braasch and Boonsoong (2010) have described new species sub nom. "*Epeorus (Iron) longitibius* Nguyen & Bae, 2004 (questionable)": 20–22 (Figs 83–98). The deposition of the female larva is in ZMKU (80% alcohol).

Adult. Unknown.

Eggs. Unknown.

Distribution. Region of Doi Inthanon (Chiang Mai province); alt 2000 m, III 1999; bottom sample (leg. R. Braasch).

Diagnosis. In contrast to the larva of *E. longitibius* with foretibiae 1.2 × length of forefemora, *E. thailandensis* has foretibia of equal length; forefemur of new species is with a small femoral spot, *E. longitibius* without such one; the gills 2-6 are quadrangular and gill 7 has a fold in *E. thailandensis*, but in *E. longitibius* gills 2-6 are elongated and gill 7 unfolded.

Habitat and biology. The larvae of *E. thailandensis* live presumably in high mountain streams with high oxygen concentrations and a faster current where the substrate is mostly stony at an elevation of 2000 m.

Remarks. Boonsoong and Braasch (2010) misidentified this species as *Iron longitibius* Nguyen & Bae, 2004 (questionable). In this study, we have re-checked them and found , then we re-identified it as a new species, *Epeorus (Iron) thailandensis*. The new species is different from the larva of *Iron longitibius* by characters within length of foretibia, femoral spot, and gill shape of gills 2–7. In this observation, we propose as the new species.

Epeorus (Epeorus) aculeatus Braasch, 1990

http://species-id.net/wiki/Epeorus_aculeatus Fig. 1A

Epeorus aculeatus Braasch, 1990: 7-9, Figs 1–8. (orig.) *Epeorus (Epeorus) aculeatus* Braasch, 1990 (comb.)

Larva. Braasch 1990: 7–9, Figs 1–8; Nguyen and Bae 2004a: 19–21, Figs 1–6. Adult. Webb and McCafferty 2006: 65–68, M, Figs 1–5. Eggs. Unknown.

Distribution. Mae Chaem district, Doi Inthanon NP (Chiang Mai province); Braasch (1990) recorded *E. aculeatus* from Doi Inthanon National Park, Chiang Mai province.

Diagnosis. The larva of *E. aculeatus* can be distinguished from that of other congeners by abdominal terga 2-9 (Fig. 1A) each bearing a pair of long, acute, submedian spines on its posterior margin, and with a median dark brown spot on femoral surface.

Habitat and biology. The larva of *E. aculeatus* occurs in headwater streams between 600–740 m alt in Thailand and high mountain streams between 1,400-2,800 m in Vietnam (Nguyen and Bae 2004a). The larvae are mostly found under rocks in fast flowing reaches of the streams. The substrate consists of mixed sand/gravel and larger stones such as boulders and cobble.

Remarks. Braasch (1990) described this species based on larval specimens. Then, Webb and McCafferty (2006) described the male imago of *E. aculeatus* based on reared Thai specimens from Doi Suthep NP, Chiangmai province. This species is different from other known species of the genus *Epeorus* by each bearing a pair of long, acute, submedian spines on its posterior margin.

Epeorus (Epeorus) bifurcatus Braasch & Soldán, 1979

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

Epeorus bifurcatus Braasch & Soldán, 1979: 266, 270, Figs 15–22. (orig.) *Epeorus (Epeorus) bifurcatus* Braasch & Soldán, 1979 (comb.)

Larva. Braasch and Soldán 1979: 266, 270, Figs 15–22; Nguyen and Bae 2004a: 21–22, Figs 7–12.

Adult. Unknown.

Eggs. Unknown.

Distribution. Tak province, highway 1090, km 64.5, mountain creek, riffle and run habitats; limy, gravel; leaf packs, wood, secondary forest; 750 m asl, c 16°30'N, 99°00'E; 14.1.2009 (leg. Freitag).

Diagnosis. The larva of *E. bifurcatus* can be distinguished from that of its congeners by pairs of small submedian dorsal tubercles on tergites with acute spines and gill 1 larger than gill 3. In the Figs 7–8 of Nguyen and Bae (2004a) some characters (the femoral spot, the rim of hairs of the hind margin of head and pronotum) are not described. Furthermore, according to our material, the head of the type species is smoothly trapezoid without markings in anterior median half, however, the head is transversely ellipsoid with a broad dark band between its front and hind margins.

Habitat and biology. Nguyen and Bae (2004a) noted that the larva of *E. bifurcatus* occur in mountain streams ranging between 200–600 m. They were found on the underside of stones in fast flowing sections of the streams.

Remarks. Braasch and Soldán (1979) described this species based on larval specimens and original descriptions are written in Germany. Then, Nguyen and Bae (2004a) described the larva of *E. bifurcatus* in English based on specimens from northern Vietnam, near the holotype locality.

Epeorus (Epeorus) inthanonensis Braasch & Boonsoong, 2010

http://species-id.net/wiki/Epeorus_inthanonensis Figs 1H–J

Epeorus inthanonensis Braasch & Boonsoong, 2010: 17–19, Figs 69–82. (orig.) *Epeorus (Epeorus) inthanonensis* Braasch & Boonsoong, 2010 (comb.)

Larva. Braasch and Boonsoong 2010: 17–19, Figs 69–82.

Adult. Unknown.

Eggs. Unknown.

Distribution. Doi Inthanon NP (Chiang Mai province).

Diagnosis. This species resembles *E. bifurcatus* however, the larva of *E. inthanonensis* has paired tubercles on the terga which are more rounded than in *E. bifurcatus*; they lack spines and gill 1 is smaller than gills 2–6. Habitat and biology. Braasch and Boonsoong (2010) noted that the larva of *E. inthanonensis* was found in small mountain streams ranging between 800–2000 m.

Remarks. This species was described by Braasch and Boonsoong (2010). This species resembles *E. bifurcatus* however, the larva has paired tubercles on the terga which are more rounded than in *E. bifurcatus*.

Genus Rhithrogena Eaton, 1881

http://species-id.net/wiki/Rhithrogena Figs 2A, 5B–D

Remarks. The genus *Rhithrogena* is the most diverse in the Holarctic, with numerous species also in the Palaearctic Asia. However, it seems to be under-represented in Southeast Asia. Braasch and Boonsoong (2009) described *R. siamensis* Braasch & Boonsoong, 2009 from northern Thailand, this species also occurs in the northeastern and western parts of Thailand. Only two further species of the genus can be found in this area: *Rhithrogena parva* Ulmer, 1939 from Taiwan (Ulmer 1912) and *Rhithrogena diehliana* Braasch & Soldán, 1986 from Sumatra (Braasch and Soldán 1986c).

Rhithrogena (Tumungula) siamensis Braasch & Boonsoong, 2009 http://species-id.net/wiki/Rhithrogena_siamensis Figs 2A, 5C–D

Rhithrogena (Tumungula) siamensis Braasch & Boonsoong, 2009: 39–43, Figs 32–47. (orig.)

Larva. Braasch and Boonsoong 2009: 39–43, Figs 32–47.

Adult. Braasch and Boonsoong 2009: 38-39, M, Figs 19-30; F, Fig. 31.

Eggs. General shape ovoid. One of the poles terminates with large knob-terminated coiled threads (KCTs) (Fig. 5C), the whole chorion is covered with uniform granules and scattered with small loose KCTs, with a large micropyle on the equatorial plane (Fig. 5D).

Distribution. Mae Hong Son province, Chiang Mai province, Chiang Rai province, Loei province.

Diagnosis. The larva of *R. siamensis* resembles *Rhithrogena (Tumungula) unica* Zhou and Peters, 2004 but differs in mouthparts structure and gill 1, with *R. siamensis* being pointed-crenulate and having longer plica which are more bluntly rounded, whereas that of *R. (T.) unica* has a few angular crenulations and a shorter, more strongly rounded plica.

Habitat and biology. *Rhithrogena siamensis* larvae cling to rock surfaces in medium- to fast-flowing water. Collections over most of the year revealed that the flight



Figure 2. A Ventral view of abdomen of *R. siamensis* Braasch & Boonsoong, 2009 **B–E** lamellae of gills 1 (**B**), 3 (**C**), 5 (**D**) and 7 (**E**) of *T. maxillaris* Braasch & Soldán, 1988 **F** ventral view of left maxilla of *T. maxillaris* Braasch & Soldán, 1988 **G** bristles on dorsal face of abdominal terga of *T. maxillaris* Braasch & Soldán, 1988 **H** ventral view of left maxilla of *C. langensis* Braasch & Boonsoong, 2010 **I-J** abdominal terga (**I**) and tergum VII (**J**) of *N. baei* Braasch & Boonsoong, 2009.

season at altitudes of 600 m was mainly during March/April, just before the beginning of the monsoon rains in May.

Remarks. Only one species of *Rhithrogena* was identified in our study as being distributed thoughout Thailand. They live in a rapid current of stream.

Genus *Trichogenia* Braasch & Soldán, 1988 http://species-id.net/wiki/Trichogenia

Figs 2B–G

Remarks. The Southeast Asian genus *Trichogenia* was established by Braasch and Soldán (1988) from Vietnam. Four species of *Trichogenia* are present in the Oriental region (Webb et al. 2006). Only one species, so far, is found in northern and northeastern Thai streams.

Trichogenia maxillaris Braasch & Soldán, 1988

http://species-id.net/wiki/Trichogenia_maxillaris Figs 2B–G

Trichogenia maxillaris Braasch & Soldán, 1988: 119-124, Figs 1–13. (orig.) *Heptagenia maxillaris* Kluge, 2004: 173. (comb.)

Larva. Braasch and Soldán 1988: 119-124, Figs 1–13.

Adult. Unknown.

Eggs. Unknown.

Distribution. Doi Suthep, Chiang Mai province; Loei province; Soppong, Mae Hong Son province.

Diagnosis. *Trichogenia maxillaris* can be differentiated from congeners by the following combination of characteristics: gill lamellae 2–7, long, narrow and sharply pointed; base of outer canines of mandibles without dense lateral brush of setae; supracoxal sclerites short. The other *Trichogenia* species have gill lamellae 5-7 rounded with a pointed apex, base of outer canines of mandibles with dense lateral brush of setae, and supracoxal sclerites long and pointed.

Habitat and biology. *Trichogenia maxillaris* larvae occur in small mountain streams. To date, only a few *Trichogenia* specimens have been collected in Thai streams. This species appears to be a sensitive indicator because larvae were found exclusively in forest stream areas.

Remarks. Only one species of *Trichogenia* (*T. maxillaries*) was reported from Thailand. The larva of this species was described by Braasch and Soldán (1988). The adults are unknown.

Genus Notacanthurus Tshernova, 1974

http://species-id.net/wiki/Notacanthurus Figs 2I–J, 6A–B

Remarks. Three species of *Notacanthurus* are described from the Himalayas (Braasch 1980, 1986). The Thai species *Notacanthurus baei* Braasch & Boonsoong, 2009 was collected and described from the northern part of country.

Notacanthurus baei Braasch & Boonsoong, 2009

http://species-id.net/wiki/Notacanthurus_baei Figs 2I–J, 6A–B

Notacanthurus baei Braasch & Boonsoong, 2009: 34-38, Figs 1-18. (orig.)

Larva. Braasch and Boonsoong 2009: 34–38, Figs 1–18.
Adult. Unknown. Its prospective penis (Fig. 14, Braasch 1986) is quite unlike the bilobed penes of the Himalayan *Notocanthurus* (Figs 4-8, Braasch 1986).

Eggs. Chorionic pattern of geometrically arranged small KCTs covering the entire egg surface (Fig. 6A) and interspersed among crenulated granules, folded surface of the chorion, many microgranules densely scattered all over the surface of the chorion, large micropyles on equatorial plane (Fig. 6B).

Distribution. Mae Hong Son province, Mae Chaem district, Doi Inthanon NP, Doi Suthep NP (Chiang Mai province)

Diagnosis. Larvae of *N. baei* are easily identified by having a dorsal median abdominal ridge on tergites 1-9 and denticles on the claws. Larvae of the Indian species *Notocanthurus edentatus* Braasch, 1986 have no dorsal ridges on the abdomen (Braasch 1986); however, all Himalayan species of *Notacanthurus* key out by the absence of denticles on the claws. Its prospective penis (Fig. 14, Braasch 1986) is quite unlike the bilobed penes of the Himalayan *Nothacanthurus* (Figs 4–8, Braasch 1986)" and is expected to be a simple, not bilobed, penis in the male imago.

Habitat and biology. *Nothacanthurus baei* larvae inhabit small streams and brooks. Larvae were usually found together with those of *Asionurus* species.

Remarks. Braasch and Boonsoong (2009) described only one species of *Nothacanthurus* from Thailand. The adults of *Nothacanthurus baei* are unknown.

Genus Thalerosphyrus Eaton, 1881

http://species-id.net/wiki/Thalerosphyrus Figs 3B–D, 6C–D

Remarks. *Thalerosphyrus* occurs from China through Southeast Asia to India. This genus is originally described from tropical Southeast Asia. *Thalerosphyrus sinuosus* Navás, 1933 seems to be widely distributed in Thailand and adjacent countries (Nguyen and Bae 2004b). Larvae of *Thalerosphyrus* are usually the dominant heptageniids in Thai streams as well as those of *Afronurus*.

Thalerosphyrus sinuosus Navás, 1933

http://species-id.net/wiki/Thalerosphyrus_sinuosus Figs 3B–D, 6C–D, 9F

Thalerosphyrus sinuosus Navás, 1933: 70, F, Fig. 80. (orig.)

Larva. Braasch and Soldán 1984b: 203–205 (sub name *Thalerosphyrus siamensis* Dang, 1967, Figs 5–20v, 26)

Adult. Navás 1933: 70, F, fig. 80; Ulmer 1939: 551–555, M&F, Figs 118–128; Braasch and Soldán 1984b: 201–206, Figs 1–8, M, 25.



Figure 3. A Right side of thorax of *A. namnaoensis* Braasch & Boonsoong, 2010 **B–D** right side of thorax (**B**), ventral view of head capsule (**C**) and ventral view of posterior abdomen (**D**) of *T. sinuosus* Navás, 1933 **E–H** lamella of gills 3 (**E**), 4 (**F**), 5 (**G**) ,6 (**H**) of *C. (Siamoneuria) kovaci* Braasch, 2006 **I–J** dorsal view of abdomen (**I**), lamella of gills 7 (**J**) of *C. thienemanni* Ulmer, 1939 **K–L** dorsal view of abdomen (**K**), lamella of gills 7 (**L**) of *C. langensis* Braasch & Boonsoong, 2010.

Eggs. KCTs randomly scattered laterally and concentrated at both poles, although larger and numerous at the pole (Fig. 6C). Rounded tubercles are scattered all over the surface of the chorion (Fig. 6D). Five – six micropyles located on equatorial plane. **Distribution.** Widely distributed in many parts of Thai streams.

Diagnosis. The combination of having a distinctly thickened anterior margin of the head capsule, long posterolateral spines on the abdomen, acutely pointed supracoxal spurs, and well-developed lamellae on gills 1 will distinguish *Thalerosphyrus* from other Ecdyonurinae genera. Within *Thalerosphyrus* is a '*sinuosus*' group of species with larvae having the above-mentioned combination, having in both sexes of adults "costal and subcostal fields with two indistict umbra-brown spots, the first at the beginning of pter-ostigmatic region, the second directly to the wing tip" (Navás 1933) and a '*determinatus*' group, having larvae with short posterolateral spines; however, both sexes of adults have umbra-brown tinged costal and subcostal fields on forewings. In general, the species differentiation in the genus (*T. 'sinuosus'* group) is unsatisfying and requires verification. The larva of *T. sinuosus* can be differentiated by the combination of the following characters: gill 3 of rounded shape is much less wide than that of *T. vietnamensis* (Dang, 1967)

and while in *T. flowersi* (Venkataraman and Sivaramakrishnan, 1987) the inner side of gill 1 has a straight margin, that of *T. vietnamensis* (=*T. sinuosus*?) is slightly concave.

Habitat and biology. *T. sinuosus* larvae (Fig. 9F) are one of the most widespread mayfly species in Thailand. The larvae are found underneath stones in slow-flowing reaches of streams (water velocity approximately 3-7 cm/sec, water depth ranges between 7-17 cm). The larvae cling to submerged boulders and cobbles. Because they feed by grazing on diatoms, algae and detritus on stream rocks, they prefer rocky substrates in fairly clear to silty sediments.

Remarks. Only one species of *Thalerosphyrus* (*T. sinuosus*) was identified from Thailand and this species widely distributed in Thai streams. The larva and adults of this species were adequately described by Braasch and Soldán (1984b) and Navás (1933).

Genus Compsoneuria Eaton, 1881

http://species-id.net/wiki/Compsoneuria Figs 2H, 3E–L, 7A–B, 9C

Remarks. The heptageniid mayfly genus *Compsoneuria* was reviewed by Webb et al. (2006). Eleven species were revised from Afro-tropical and Oriental regions. *Compsoneuria* larvae were found abundantly in lowland rivers on mainland Southeast Asia. There are three species of *Compsoneuria* mayflies reported from Thailand (Braasch 1990; Braasch 2006a; Braasch and Boonsoong 2010). *Compsoneuria* larvae occur among submerged vascular plants or roots.

Compsoneuria thienemanni Ulmer, 1939

http://species-id.net/wiki/Compsoneuria_thienemanni Figs 3I–J, 7A–B, 9C

Compsoneuria thienemanni Ulmer, 1939: 672, Figs 440–448, 449–454. (orig.) Compsoneuria thienemanni Braasch and Soldán, 1986b: 46. (comb.) Thalerosphyrus thienemanni Wang and McCafferty, 2004: 17. (comb.)

Larva. Ulmer 1939: 672, Figs 440–448, 449–454; Braasch and Soldán 1986b: 42–46, Figs 14.1–14.14.

Adult. Ulmer 1939: 564, M, Figs 145–149, 152; F, 151; Fs, 150; Braasch and Soldán 1986b: 42–44, M, Figs 5-9.

Eggs. Chorionic surface characterized by granular matrix, large KCTs randomly scattered on entire egg surface (Fig. 7A), small micropyles visible on equatorial plane (Fig. 7B).

Distribution. Mae Hong Son province, Chiang Mai province, Trat province.

Diagnosis. *Compsoneuria thienemanni* is recognised by having gills 2-6 without emarginations and gill 7 leaf-like and pointed apically; pale dots and marks on abdomen as shown in Fig. 3I.

Habitat and biology. *Compsoneuria thienemanni* larvae (Fig. 9C) are found abundantly in the large rivers at lower altitudes where it is encountered clinging to floating submerged water plants. A few male specimens of *C. thienemanni* were found in the mountainous region of Mae Hong Son province.

Remarks. The larva and adults of this species were described by Ulmer (1939) and Braasch and Soldán (1986b). The larvae of this species are found abundantly in the low-gradient streams and large rivers in Thailand.

Compsoneuria langensis Braasch & Boonsoong, 2010

http://species-id.net/wiki/Compsoneuria_langensis Figs 2H, 3K–L

Compsoneuria langensis Braasch & Boonsoong, 2010: 9-11, Figs 31-45. (orig.)

Larva. Braasch and Boonsoong 2010: 9–11, Figs 31–45.

Adult. Braasch and Boonsoong 2010: 7–9, M, Figs 19, 21–25, 27–30; F, Figs 20, 26. **Eggs.** Unknown.

Distribution. Nam Lang River, Soppong (Mae Hong Son province).

Diagnosis. Larva without emarginations on gills 2–6, gill 7 lanceolate and rounded apically, pale dots and marks on abdomen as on Fig. 3K.

Habitat and biology. Both *C. thienemanni* and *C. langensis* larvae are mainly found attached to floating water plants or thick pads of green algae.

Remarks. Braasch and Boonsoong (2010) described this species from Thailand based on larval and imaginal specimens. The larva of this species is different from other known species by gills 2–6 without emarginations, gill 7 lanceolate and rounded apically.

Compsoneuria (Siamoneuria) kovaci Braasch, 2006

http://species-id.net/wiki/Compsoneuria_kovaci Figs 3E–H

Compsoneuria (Siamoneuria) kovaci Braasch, 2006: 50-51, Figs 9-21. (orig.)

Remarks. A brief comment on the appropriate placement of *C. (S.) kovaci* in Braasch and Boonsoong (2010) is required. Larval diagnosis of *Compsoneuria (Siamoneuria)* is in contradiction to that of *Compsoneuria*, namely in lacking the combination of long, sharply pointed supracoxal spurs, black spotting on the head capsule and femora, and narrow, apically pointed glossae. *Siamoneuria* cannot belong in the genus *Compsoneuria* but probably deserves its own status; it appears to be a morphospecies whose characters are not in accordance with other known genera. For a final definition, important missing details of mouthparts and eggs should be included. For now, we see it as a

species as "INCERTAE SEDIS" (Braasch and Boonsoong 2010). So, further studies and more specimens are needed to clear taxonomic of this species.

Larva. Braasch 2006a: 50–51, Figs 9–21.

Adult. Braasch 2006a: 49–51, M, Figs 1–8.

Eggs. Unknown.

Distribution. Nam Lang river, Soppong, Mae Hong Son province,

Diagnosis. Larva is conspicuous due to emarginations on gills 2–6, gill 7 being narrowly lanceolate, the head without markings, and the body with paired paramedian spots on tergites 5–8, large median spot on 9, and three distantly arranged small spots at anterior margin of tergite 10.

Habitat and biology. The habitat of the single larva found was submerged roots of a tree standing at the river bank.

Genus Asionurus Braasch & Soldán, 1986

http://species-id.net/wiki/Asionurus Figs 4A–B, 7C–D, 9B

Remarks. Three species of *Asionurus* mayflies have been reported from the Oriental region (Braasch 2011; Braasch and Soldán 1986a; Braasch and Soldán 1986b). Only *A. primus* Braasch & Soldán, 1986 was collected and reported from northern Thailand. The identity of Vietnamese specimens of *A. primus* (Braasch and Soldán 1986a) with those of northern Thailand (Sangpradub et al. 2002) is probable but needs confirmation by reared males from Vietnam.

Asionurus primus Braasch & Soldán, 1986

http://species-id.net/wiki/Asionurus_primus Figs 4A–B, 7C–D, 9B

Asionurus primus Braasch & Soldán, 1986a: 155–158, Figs 1–13. (orig.)

Larva. Braasch and Soldán 1986a: 155–158, Figs 1–13.

Adult. Braasch and Boonsoong 2010: 5–7, M, Figs 14–17; F, Fig. 18.

Eggs. General shape ovoid, both poles with large KCTs densely arranged (Fig. 7C), many microgranules densely scattered all over the surface of the chorion, macrogranules on equatorial plane (Fig. 7D), border well-defined by a thickened rim beset with tubercles, 5–6 micropyles on equatorial plane.

Distribution. Chaiyaphum province, Mae Hong Son province, Chiang Mai province.

Diagnosis. *A. primus* differs from *A. ulmeri* (Braasch and Soldán 1986a) by shorter and more pointed wings of hypopharynx and gill 7 with bulging anterior portion and acutely shaped apically. In *A. ulmeri* the wings of hypopharynx are longer and have rounded ends whereas gill 7 is narrow and long and hardly extended anterioriorly.



Figure 4. A–B Lamella of gills 7 (A) and setae on inner surface of hind tarsi (B) of *A. primus* Braasch & Soldán, 1986 C bristles on cerci of *R. tonkinensis* Soldán & Braasch, 1986 D bristles on cerci of *A. nam-naoensis* Braasch & Boonsoong, 2010 E dorsal view of abdomen of *A. rubromaculatus* You, Wu, Gui & Hsu, 1981 F lamella of gills 1 of *A. gilliesiana* Braasch, 1990 G lamella of gills 7 of *A. rainulfiana* Braasch, 1990.

Habitat and biology. *A. primus* larvae (Fig. 9B) are often the most abundant in small mountain streams of Thailand. Larvae live beneath rocks and debris. They were found together mostly with those of *Notacanthurus baei*. Larval habitat preference is similar to that of larvae of *N. baei*.

Remarks. Only one species of *Asionurus (A. primus)* was identified from Thailand. The larva and adults of this species was described by Braasch and Soldán (1986a) and Braasch and Boonsoong (2010). The larva of this species found in small mountain streams of Thailand.

Genus *Rhithrogeniella* **Ulmer, 1939** http://species-id.net/wiki/Rhithrogeniella Figs 4C, 8A–B, 9E

Remarks. Genus *Rhithrogeniella* is unique in having scaled caudal filaments with special arrangements of stout bristles and finer setae in the proximal portion; distal portion segments bear longer, stiffer setae and at articulations short bristles alternate with fine



Figure 5. A–B General outline (**A**) and micropyle (**B**) of the egg of *E. khayengensis* Boonsoong & Braasch, 2010 **C-D** General outline (**C**) and micropyle (**D**) of the egg of *R. siamensis* Braasch & Boonsoong, 2009. Scale bars 20 μ m for **A** and **C**; 5 μ m for **B** and **D**.

setae. These characters are similarly expressed in several *Nixe* spp. from Taiwan (Kang and Yang 1994). In view of the similarity in egg structure *Rhithrogeniella* is recently often identified as *Nixe* Flowers, 1980. However, Ulmer(1939) published *Rhithrogeniella* ornata from Sundaland which clearly has priority over *Nixe* Flowers, 1980.



Figure 6. A–B General outline (**A**) and micropyle (**B**) of the egg of *N. baei* Braasch & Boonsoong, 2009 **C-D** General outline (**C**) and micropyle (**D**) of the egg of *T. sinuosus* Navás, 1933. Scale bars 20 μ m for **A** and **C**; 5 μ m for **B** and **D**.



Figure 7. A–B General outline (**A**) and micropyle (**B**) of the egg of *C. thienemanni* Ulmer, 1939 **C–D** General outline (**C**) and micropyle (**D**) of the egg of *A. primus* Braasch & Soldán, 1986. Scale bars 20 μ m for **A** and **C**; 5 μ m for **B** and **D**.



Figure 8. A–B General outline (**A**) and micropyle (**B**) of the egg of *R. tonkinensis* Soldán & Braasch, 1986 **C–D** General outline (**C**) and micropyle (**D**) of the egg of *A. namnaoensis* Braasch & Boonsoong, 2010. Scale bars 20 µm for **A** and **C**; 5 µm for **B** and **D**.



Figure 9. A Habitus of *A. namnaoensis* Braasch & Boonsoong, 2010 B habitus of *A. primus* Braasch & Soldán, 1986 C habitus of *C. thienemanni* Ulmer, 1939 D habitus of *E. khayengensis* Boonsoong & Braasch, 2010 E habitus of *R. tonkinensis* Soldán & Braasch, 1986 F habitus of *T. sinuosus* Navás, 1933.

Rhithrogeniella ornata Ulmer, 1939

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

Rhithrogeniella ornata Ulmer, 1939: 575-576, Figs 165-174. (orig.)

Adult. Ulmer 1939: 575–576, M, Figs 165–166, 169; Ms, fig. 170, 173–174; F, Figs 171–172; Fs, Figs 167–168.

Rhithrogeniella tonkinensis Soldán & Braasch, 1986

http://species-id.net/wiki/Rhithrogeniella_tonkinensis Figs 4C, 8A–B, 9E

Rhithrogeniella tonkinensis Soldán & Braasch, 1986: 203-210, Figs 1-18. (orig.)

Larva. Soldán and Braasch 1986: 203–210, Figs 1–18.

Adult. Soldán and Braasch 1986: 203 (F), 206, 210 (Ms, Figs 19–22); Braasch 1990: 11–12, M, Figs 17.1–17.4.

Eggs. Egg ovoid, chorionic surface with mesh-like reticular ridges of a hexagonal structure, evenly covered with KCTs (Fig. 8A), micropyle slightly oval with inconspicuous marginal rim, 1–2 small micropyles visible on the equatorial area (Fig. 8B).

Distribution. Chiang Mai province, Chaiyaphum province.

Diagnosis. The larvae of *R. tonkinensis* can be distinguished from those of other genera of Heptageniidae by the following combination of characters: the head is approximately as broad as the pronotum, without a median emargination and marginal bristles and by the presence of interfacing setae on the caudal filaments (Soldán and Braasch 1986). The latter are provided at rings with rather stout spines regularly alternating with fine setae. Segments of filaments are "scaled". Larvae of the other Southeast Asian species *Rhithrogeniella ornata* Ulmer, 1939 are unknown.

Habitat and biology. Larvae of *R. tonkinensis* (Fig. 9E) occur in relative deep waters (30-40 cm) with slow currents and smaller stones or coarse sand on the bottom. Larvae are good swimmers, but prefer to remain attached to the stone surface rather than swimming (Soldán and Braasch 1986).

Remarks. The larva and adults of *R. tonkinensis* were adequately described by Soldán and Braasch (1986). Only *R. tonkinensis* found in relative deep waters with slow currents of Thai streams.

Genus Afronurus Lestage, 1924

http://species-id.net/wiki/Afronurus Figs 3A, 4D–G, 8C–D, 9A

Remarks. The synonymization of *Cinygmina* with *Afronurus* is recognized by Wang and McCafferty (2004), Kluge (2004), and Braasch and Freitag (2008). The genus *Afronurus* includes at least 43 species from the Oriental region (Braasch 1987, 1990, 2005, 2011; Braasch and Boonsoong 2010, Braasch and Soldán 1984a, 1987, Flowers and Pescador 1984, Braasch and Jacobus 2011, Kang and Yang 1994, Kimmins 1937, Nguyen and Bae 2003, Venkataraman and Sivaramakrishnan 1989, Zhou and Zheng 2003). It indicates the complicated situation in determining species of *Afronurus* in the Oriental; in Southeast Asia many species are known only by larvae, or described as adults with affiliation of larvae from the same locality; the rearing of

species and genetic investigations will be the aim of a future research. *Afronurus* larvae are usually the dominant heptageniid benthic macro-invertebrates in Thai streams. In this study, we propose five described species of the genus *Cinygmina* Kimmins, 1937 = *Afronurus* Lestage, 1924.

Afronurus cervina Braasch & Soldán, 1984

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

Cinygmina cervina Braasch & Soldán, 1984: 196–197, 199, Figs 17–31. (orig.) *Afronurus cervina* Braasch & Soldán, 1984 (comb.)

Larva. Braasch and Soldán 1984a: 196–197, 199, Figs 17–31, Vietnam; no record in Thailand.

Adult. Braasch and Soldán 1984a: Vietnam; 196–197, 199, M, Figs 14–16; Braasch 1990: 8, Fs & Ms, Thailand.

Eggs. Unknown.

Distribution. Ban Nam Tok (Chiang Rai province).

Diagnosis. Braasch (1990) reported this species based on male, female and subimago male and female specimens, and the head of a presumed larva of *A. cervina* without markings; gill 1 somewhat upturned, narrowly banana-shaped (Figs 17–18, Braasch and Soldán 1984).

Habitat and biology. This species is found to be an inhabitant of fast flowing rivers in Vietnam.

Remarks. The larva and adults of *A. cervina* were described by Braasch and Soldán (1984a). Only adults of *A. cervina* found in Thailand. But, the larva of this species is not found in Thai streams.

Afronurus dama Braasch & Soldán, 1987

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

Cinygmina dama Braasch & Soldán, 1987: 125, Figs 7.1–7.4. (orig.) *Afronurus dama* Braasch & Soldán, 1987 (comb.)

Larva. Braasch and Soldán, 1987: Vietnam; 125, Figs 7.1–7.4.

Adult. Braasch and Soldán 1987: 123, 125, 126, M, Figs 8.1–8.3; Braasch 1990:

8 (reported 2 M and 1 F from Thailand).

Eggs. Unknown.

Distribution. Nam Tok Ban Du (Chiang Rai province).

Diagnosis. Head with blurred spots at forward margin; gill 1 up-turned bananashape, 3 smoothly triangular gills with obliquely attached projection. **Habitat and biology.** The larvae of *A. dama* were found in streams of Tam Dao, Song Dan, Vinh Puh province, Vietnam.

Remarks. Only adults of *A. dama* were reported from Thailand (Braasch 1990). The larva and adults of *A. dama* were described by Braasch and Soldán (1987).

Afronurus gilliesiana Braasch, 1990

http://species-id.net/wiki/Afronurus_gilliesiana Fig. 4F

Cinygmina gilliesiana Braasch, 1990: 8, 10, Figs 13.1–13.4, 14–16. (orig.) *Afronurus gilliesiana* Braasch, 1990 (comb.)

Larva (F). Braasch 1990: 8, 10, Figs 13.1–13.4, 14–16.

Adult. Unknown.

Eggs. Unknown.

Distribution. Mae Sot district (Tak province).

Diagnosis. The larva of *A. gilliesiana* can be distinguished from congeners by the combination of the following characters: head with indistinct spots; gill 1 broadly lanceolate (Fig. 13.1, Braasch 1990); gill 3 widely rounded triangular (Fig. 13.2, Braasch 1990), gill 5 obliquely rounded triangular with small projection (Fig. 13.3, Braasch 1990), and broad asymmetrically oval gill 7 (Fig. 13.4, Braasch 1990).

Habitat and biology. The larvae of *A. gilliesiana* were found in headwater streams in northern Thailand.

Remarks. Only larva of *A. gilliesiana* was reported from northern part of Thailand (Braasch 1990). The larva of *A. gilliesiana* were described by Braasch (1990). The adults of *A. gilliesiana* are unknown.

Afronurus namnaoensis Braasch & Boonsoong, 2010

http://species-id.net/wiki/Afronurus_namnaoensis Figs 3A, 4D, 8C–D, 9A

Afronurus namnaoensis Braasch & Boonsoong, 2010: 1-3, Figs 6-13. (orig.)

Larva. Braasch and Boonsoong 2010: 1–3, Figs 6–13.

Adult. Braasch and Boonsoong 2010: 2–3, M, Figs 1–4; F, fig. 5.

Eggs. The egg chorion of *A. namnaoensis* is decorated with granules and two kinds of KCTs: small KCTs concentrated at each pole and much larger oval KCTs located equatorially (Fig. 8C); micropyles have an ovoid to round sperm guide (Fig. 8D), visible in the equatorial area. The micropyle is interposed between adjacent equatorial KCTs.

Distribution. Phromlaeng stream (Chaiyaphum province); Yakraue stream (Petchabun province); Nam Lang river, Pangmapa/Soppong (Mae Hong Son province); Chiang Mai province.

Diagnosis. Male of *A. namnaoensis* is separated from Vietnamese *A. cervina* in lacking a median penial cone, by the less deeply notched lobal apex, and titillators curved laterally and in their more medial position. Vietnamese *A. dama* presents a terminal apex of the penis slightly notched at the inner angles, whereas *A. namnaoensis* is recognized by somewhat elevated corners on both sides of the apices. Larvae are recognizable by an unmarked forehead, pointed and weakly curved gill 1 and smoothly rounded triangular gill 5 with crosswise projection. Two Vietnamese species, *Afronurus meo* and *A. mnong* (Nguyen and Bae, 2003) have gills lacking these projections (Figs 8–10, Figs 18–20, Nguyen and Bae 2003).

Habitat and biology. Larvae of *A. namnaoensis* (Fig. 9A) are probably the most abundant species on rocks and stones in Nam Lang River and elsewhere in current waters of northern and northeastern Thailand. These mayflies are an important food source for headwater stream fishes (*Cyclocheilichthys apogon, Devario regina, Opsarius pulchellus,* and *Rasbora rasbora*).

Remarks. Braasch and Boonsoong (2010) described this species from Thailand based on nymphal and imaginal specimens, and deposited them in the ZMKU, Bangkok. The larvae of *A. namnaoensis* are the most abundant species in current waters of Thai streams.

Afronurus rainulfiana Braasch, 1990

http://species-id.net/wiki/Afronurus_rainulfiana Fig. 4G

Cinygmina rainulfiana Braasch, 1990: 8, 10, 11, Figs 9–12, Figs 18.1–18.3. (orig.) *Afronurus rainulfiana* Braasch, 1990 (comb.)

Larva (M). Braasch 1990: 8, 10, 11, Figs 9–12, Figs 18.1–18.3.

Adult. Unknown.

Eggs. Unknown.

Distribution. Mae Sot district (Tak province).

Diagnosis. The larvae of *A. rainulfiana* can be distinguished from congeners by the combination of the following characters: head with a distinct pattern of light spots (Fig. 9), a broad banana-shaped gill 1 (Fig. 18.1, Braasch 1990), asymmetrically oval gill 6 with sloping finger-like projection (Fig. 18.2, Braasch 1990) and gill 7 narrowly lanceolate (Fig. 18.3, Braasch 1990).

Habitat and biology. The larvae of A. rainulfiana were found in headwater streams.

Remarks. Only larva of *A. rainulfiana* was reported from northern part of Thailand and was described by Braasch (1990). The adults of *A. rainulfiana* are unknown.

Afronurus rubromaculata You, Wu, Gui & Hsu, 1981

http://species-id.net/wiki/Afronurus_rubromaculata Fig. 4E

Cinygmina rubromaculata You, Wu, Gui & Hsu, 1981: 4, Figs 1–13. (orig.) *Afronurus rubromaculata* You, Wu, Gui & Hsu, 1981 (comb.)

Larva. Wu et al. 1986: 67, Figs 1–10; Zhou and Zheng 2003: 757, Figs 7–10. Adult. You et al. 1981: 4, M & F, Figs 1–13; Zhou and Zheng 2003: 758, Fig. 17.

Eggs. Unknown.

Distribution. Ban Nam Tok (Chiang Rai province); Nam Lang river, Soppong, Mae Hong Son province.

Diagnosis. This species is unique in the genus because of its abdominal pigmentation: terga pale yellow medially and reddish laterally. The male genitalia have an obvious projection between the two lobes. The larvae of this species are larger and have more pale dots and marks on head and body than those of the other known species (Figs 7, 9, 10, Zhou and Zheng 2003), gill 5 or 6 are provided with a small, thin projection (Fig. 8).

Habitat and biology. Larvae of *A. rubromaculata* were the only representatives of *Afronurus* encountered in the large river Mekong in February 2002 along the Thai-Laos border in the utmost north of Thailand. It is regularly found as a resident together with the dominant *A. namnaoensis* on stones and rocks in Nam Lang River, altitude 600 m (Braasch 2006). This species is also found in Vietnam.

Remarks. The larva and adults of *A. rubromaculata* were adequately described by You et al. (1981), Wu et al. (1986) and Zhou and Zheng (2003). Only larva of *A. rubromaculata* was reported in Thailand (Braasch and Boonsoong 2010).

A key to the eggs of known genera and species of Heptageniidae in Thailand

1	KCTs absent (Fig. 5A) E. khayengensis Boonsoong & Braasch
-	KCTs present
2	Small KCTs densely concentrated at each pole, much larger KCTs equatori-
	ally (Fig. 8C)
-	KCTs not as above
3	Chorion tuberculate or with peg-like structures
_	Chorion reticulate (Fig. 8A)
4	Coils concentrated at one or both poles and evenly distributed about remain-
	der of egg (Fig. 5C, 6C, 7C)
-	Coils never concentrated at poles; evenly distributed around entire egg (Fig.
	6A, 7A)

5	Coils concentrated at one pole; chorion surface with peg-like structure (Fig.
	5C)
-	Coils concentrated at both poles; chorion surface tuberculate
6	Many microgranules densely scattered all over the surface of the chorion;
	micropyle border well defined by a thickened rim beset with tubercles (Fig.
	7C)A. primus Braasch & Soldán
-	Rounded tubercles are scattered all over the surface of the chorion; micropyle
	border not strongly thickened (Fig. 6C) T. sinuosus Navás
7	Chorionic surface folded, with many densely scattered crenulated granules;
	small KCTs covering the entire egg surface (Fig. 6A)
	N. baei Braasch & Boonsoong
-	Chorionic surface with many sizes of granular matrix; large KCTs randomly
	scattered (Fig. 7A) C. thienemanni Ulmer

Conclusions and recommendations

Heptageniidae is the most diverse and abundant mayfly family in Thailand. The total number of Thai Heptageniidae described to date amounts to 9 genera and 22 species. The results presented here show that the Thai heptageniid fauna is dominated by Oriental genera. In addition, three Southeast Asian endemic genera morphospecies (Asionurus, Siamoneuria and Trichogenia) are found in Thailand. The species of the genera Afronurus and *Thalerosphyrus* are the most abundantly and widely distributed species found in Thai streams. Early studies of Thai heptageniid mayflies provide an important base on which to continue the study of these insects. It is important, now, to investigate the fauna further, which will lead to a better biogeographical understanding, and, at the same time, to begin a study of the biology and ecology, which is still very limited for species recorded in Thailand. Data on the ecology of heptageniids in Asian streams is limited. Dudgeon (1996) gives some preliminary data on life histories of five Hong Kong species, and estimates of their secondary production. Boonsoong (2002) presents some preliminary data on life history and diet of Afronurus species from Nam Nao National Park. These larvae show a non-seasonal multivoltine. Based on gut analyses, larvae are non-selective generalists. They feed mainly on detritus and diatoms and could be categorized as scrapers. Larvae of heptageniid mayflies often occur in reaches of fast-flowing streams where mixed substrates are composed of cobble, pebble, and gravel. In general, the distribution of heptageniid larvae depends upon substrate type and water current.

In addition to basic taxonomic research, revision of unclear or poorly defined genera, and association of larval and adult stages by rearing, investigation priorities of the Thai Heptageniidae can be summarized briefly as follows: study of life cycle and ecology of individual species distribution; of heptageniid larvae with respect to different water conditions.

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