RESEARCH ARTICLE



# The first records of Stenobermuda Schultz, 1982 and Tenupedunculus Schultz, 1979 from Australia, with description of two new species from the Great Barrier Reef (Isopoda, Asellota, Stenetriidae)

Ji-Hun Song<sup>1,\*</sup>, Niel L. Bruce<sup>2,3,\*</sup>, Gi-Sik Min<sup>1</sup>

 Department of Biological Sciences, Inha University, 100 Inha-ro, Nam-gu, Incheon 22212, South Korea
Museum of Tropical Queensland, Queensland Museum, 70-102 Flinders Street, Townsville, Australia 4810
Unit for Environmental Sciences and Management Water Research Group (Ecology), North West University, Potchefstroom 2520, South Africa

Corresponding author: Gi-Sik Min (mingisik@inha.ac.kr)

Academic editor: S. Sfenthourakis   Received 21 August 2017   Accepted 1 December 2017   Published 26 January 2018
http://zoobank.org/6CDB4835-761A-4D11-8C53-E4C066A8E563

**Citation:** Song J-H, Bruce NL, Min G-S (2018) The first records of *Stenobermuda* Schultz, 1982 and *Tenupedunculus* Schultz, 1979 from Australia, with description of two new species from the Great Barrier Reef (Isopoda, Asellota, Stenetriidae). ZooKeys 733: 1–24. https://doi.org/10.3897/zookeys.733.20474

# Abstract

The genera *Tenupedunculus* Schultz, 1982 and *Stenobermuda* Schultz, 1979 are recorded for the first time from beyond the Southern Ocean, at the Great Barrier Reef, Australia. *Tenupedunculus serrulatus* **sp. n.** and *Stenobermuda warooga* **sp. n.** are described from Heron Island and Lizard Island respectively, both in the Great Barrier Reef. The genus *Tenupedunculus* is revised and a new diagnosis presented, with *Tenupedunculus virginale* Schultz, 1982, *T. pulchrum* (Schultz, 1982), and *T. serrulatus* **sp. n.** being retained within the genus, and the remaining species here regarded as Stenetriidae *incertae sedis* (eight species).

# Keywords

Asellota, Australia, Great Barrier Reef, new species, Stenetriidae, Stenobermuda, Tenupedunculus

\* Equal contributors

# Introduction

The family Stenetriidae Hansen, 1905 comprises 12 accepted genera (Wilson and Schotte 2008). Serov and Wilson (1995) provided the only comprehensive generic review and reappraisal of this family. More recently the genera *Machatrium* Bruce & Buxton, 2013 and *Onychatrium* Bruce & Cumming, 2015 were described, including new species from the Great Barrier Reef, Australia. Marine stenetriids are diverse, abundant and omni-present on coral reefs (Kensley 1984a; b; 1988; Müller 1990; 1991a, b; Kensley and Schotte 2002; Martin et al. 2003), but still remains relatively poorly documented in tropical Australia (see Bruce and Cumming 2015).

In this paper, two new species of Stenetriidae are described from Great Barrier Reef, Australia: *Tenupedunculus serrulatus* sp. n. and *Stenobermuda warooga* sp. n. These are the first records of these genera from Australian waters. The genus *Stenobermuda* is known to occur on coral reefs in East Africa (Kensley and Schotte 2002) and this is the first record of the genus from the western Pacific. *Tenupedunculus* has hitherto included deep-water Southern Ocean species, often incompletely described, and presenting an inconsistent suite of characters at the generic level. *Tenupedunculus* is here revised and a restrictive diagnosis presented. *Tenupedunculus serrulatus* is the first record of the genus from shallow water and from coral reefs.

Including the present genera the Great Barrier Reef is now known to have four genera of Stenetriidae. The rich collections made during the Census of Marine Life's (CoML) Census of Coral Reef Ecosystems (CReefs) Program, housed at the Museum of Topical Queensland, hold abundant specimens of the genera *Liocoryphe* Serov & Wilson, 1995, *Tristenium* Serov & Wilson, 1995, *Mizothenar* Serov & Wilson, 1995 and *Stenetrium* Haswell, 1881 (from Ningaloo Reef, Western Australia).

#### Materials and methods

**Sampling.** See Bruce (2015) and Bruce and Buxton (2013) for details of sampling methods and locations.

**Descriptions.** See Bruce and Buxton (2013) for a detailed account of pereopod morphology. Descriptions were generated using a DELTA database (Dallwitz et al. 2006; Coleman et al. 2010). Whole animals were drawn using a stereomicroscope (Leica MZ125, Wetzlar, Germany) and dissected appendages were drawn using a light microscope (Leica DM2500, Wetzlar, Germany) equipped with differential interference contrast and a camera lucida. Dissected appendages were temporarily mounted on slides using an 85% lactic acid solution, lightly stained with lignin pink.

**Digital inking.** Pencil illustrations were scanned and electronically inked using a graphics tablet (Wacom Intuos4, Düsseldorf, Germany) and Adobe Illustrator CS5. A dorsal view of the pleotelson for each species was drawn with the aspect specifically positioned to allow for descriptive measurements. In habitus drawings, specimen curvature of these animals often distorts the true length of the pleotelson.

Permits. Specimens were collected under permits: Great Barrier Reef Maine Park Authority GBRMPA G08/26156.1, G09/32313.1; Queensland Fisheries Service QFS 95152. Abbreviations. MTQ – Museum of Tropical Queensland; RS – robust seta/e.

# Taxonomy

# Suborder Asellota Latreille, 1802 Family Stenetriidae Hansen, 1905

### Genus Tenupedunculus Schultz, 1982 sensu stricto

Tenupedunculus Schultz, 1982: 77.- Serov and Wilson 1995: 77.

**Type species.** *Tenupedunculus elongatus* Schultz, 1982; by original designation and monotypy.

**Species included.** *Tenupedunculus elongatus* (type species), south-eastern Argentine Basin, 4696 m; *T. virginale* (Schultz, 1982), Scotia Sea, Antarctica, 567 m; *T. pulchrum* (Schultz, 1982), southern Argentina, 1911 m; *T. serrulatus* sp. n., Great Barrier Reef, Australia, 25 m.

Species here excluded from *Tenupedunculus s. str.*, are regarded as **Stenetriidae** *incertae sedis*: *Tenupedunculus acutum* (Vanhöffen, 1914); *T. beddardi* (Kussakin, 1967); *T. dentimanum* (Kussakin, 1967); *T. drakensis* (Schultz, 1982); *T. inflectofrons* (Schultz, 1982); *T. serraticaudum* (Kussakin & Vasina, 1984); *T. smirnovi* (Vasina, 1982); and *T. haswelli* (Beddard, 1886).

**Diagnosis (male).** Cephalon frontal margin antennal spines small; lateral spines moderate, acute, slightly longer than antennal spines or sub-equal length. Pseudor-ostrum quadrate to trapezoid, wider than long. Eyes small, round. Male pereopod 1 ischium–carpus superodistal margin produced with acute process, inferodistal margins not produced; propodus moderate, length 1.2–1.5 times maximum width, 1.9–2.1 times carpus length, propodal palm transversely truncate or distally inflected; dactylus length similar to propodus distal width. Male pleopod 2 appendix masculina bluntly rounded apically, without apical setae.

**Description (male).** Body dorsal surface smooth or sparsely setose, widest at pereonites 6 and 7; pereonite 1 length greater than 0.9 times pereonite 2 length; pereonites 2–4 lateral margins convex, anteriorly acute. Pleotelson length subequal to width; lateral margins or finely serrate, sub-parallel, posterolateral spines prominent, margin posterior to spines rounded with weak or no apical lobe; dorsal surface smooth, or sparsely setose. Cephalon lateral margins smooth or finely serrate. Antennae length equal or longer than total body length, article 1 lateral spine absent. Pereopod 1 basis superior margin with irregularly spaced setae along length; propodal palm with teeth along palm margin; dactylus length subequal to propodal palm length. Pleopod 1 protopod rectangular, lateral margin setae present; rami lateral margins evenly convex. Pleopod 2 protopod longer than wide, distal apex blunt, transversely truncate; *appendix masculina* lateral margin groove absent. *Pleopod 5* distal apex with 3–5 plumose setae.

**Remarks.** The genus *Tenupedunculus* Schultz, 1982 was established as a monotypic genus based on a single male specimen lacking legs. Serov and Wilson (1995), who included eleven species in the genus, doubted the unity (= monophyly) of *Tenupedunculus* without referring to specific characters. Our overview of the species included in *Tenupedunculus* recognizes that many species are inadequately described, some lacking details of any pereopods or of the male pereopods and some lacking description of the male pleopods 1 and 2. There are two significant characters that differ between the species formerly placed in *Tenupedunculus* that we consider to be of generic significance, namely eye shape and pseudorostrum shape. Those species with large reniform eyes (or traces thereof) and an elongate pseudorostrum are here all excluded from *Tenupedunculus* Schultz, 1982 *sensu stricto*.

The characters that serve best to identify the genus *Tenupedunculus* are: small round eyes; anterior margin of head with only lateral spines prominent; pseudorostrum wider than long, quadrate or trapezoid; inferodistal margin of ischium–carpus in male pereopod 1 without process, and superodistal margin of ischium–carpus usually produced and acute (strongly produced as a process in *T. serrulatus* sp. n.). The principle differentiating and diagnostic characters for *Tenupedunculus sensu stricto* are presented in Table 1.

**Distribution.** All species of the genus, with the exception of the new species, are from the Antarctic and the sub-Antarctic region—off Argentina, Patagonian Shelf and also Scotia Sea (all Atlantic sector); at depths 500 to 4696 metres. *Tenupedunculus ser-rulatus* sp. n., from the southern Great Barrier Reef, is the first record of the genus from beyond the Southern Ocean and from depths less than 500 metres.

**Remarks on the species excluded from** *Tenupedunculus.* The species listed here are retained in *Tenupedunculus* Schultz, 1982, but excluded from the genus *sensu stricto* as they either lack the diagnostic characters of *Tenupedunculus* or possess unique characters that also preclude their inclusion in other stenetriid genera. The here termed '*dentimanum* group' of species, particularly when considered in conjunction with their shared characters, potentially warrants a new genus.

### 'dentimanum group'

All species share the following characters: cephalon antennal spines small, lateral spines large. Pseudorostrum anteriorly round to acute, as long as or longer than wide. Eyes large, reniform. Male pereopod 1 ischium and merus superodistal margins weakly to strongly forming an acute process, inferodistal margins not produced. Male pleopod 2 appendix masculina excavate, apically rounded, with apical setae.

*Tenupedunculus beddardi* (Kussakin, 1967). Southern Argentina; 680 m; similar to *T. dentimanum* with the following characters common to the group: pseudorostrum approximately as long as wide, lateral spines large, eyes reniform and male pereopod 1 carpus with distinct acute superodistal process. The pseudorostrum uniquely converges to a narrowly rounded apex. The appendix masculina is acute (not excavate), differing significantly from that of others in this group, but on

	Tenupedunculus sensu stricto	ʻ <i>dentimanum</i> group'	
1. Eyes	small, round	large, reniform	
2. Cephalon, lateral spines	moderate	large	
3. Pseudorostrum	quadrate to trapezoid, wider than long	round to acute, longer than wide	
4. Pereopod 1 ischium–carpus superodistal margin	produced, with acute process	produced, with acute process (except carpus)	
5. Appendix masculina	without apical setae	with apical setae	
6. Appendix masculina	bluntly rounded apically	excavate rounded apically	

Table 1. Principle differentiating characters for *Tenupedunculus sensu stricto* (male).

balance the species otherwise agrees well with and is best placed within the '*denti-manum* group' at present.

- *Tenupedunculus dentimanum* (Kussakin, 1967). Southern Argentina; 680 m; pseudorostrum as long as posterior width, anteriorly broadly rounded.
- *Tenupedunculus inflectofrons* (Schultz, 1982). Scotia Sea, Antarctica; 588 m; pseudorostrum rounded; male pereopod 1 not known; appendix masculina with terminal process.
- *Tenupedunculus smirnovi* (Vasina, 1982). Patagonian Shelf; 500 m. Female only; seems to have reniform eyes, pseudorostrum stepped, acute; female pereopod 1 with ischium and merus with acute superodistal margin but not carpus; pleotelson with distinct caudomedial lobe.

# Ungrouped species.

*Tenupedunculus acutum* (Vanhöffen, 1914). Gauss Station, Davis Sea; 3397 m; pseudorostrum longer than wide, anteriorly rounded with median point; moderate lateral spines on cephalon; eyes moderate in size (more than six ommatidia) round (eye shape is not entirely clear in the original figures); appendix masculina blunt (excavate), with apical setae; male pereopod 1 ischium and merus with acute processes but carpus without process. Eye size and shape precludes inclusion of *T. acutum* in the '*dentimanum* group'.

- *Tenupedunculus haswelli* (Beddard, 1886). Rio del la Plata; 1097 m; eyes reniform; male pereopod 1 with superodistal process on carpus (i.e. pereopod 1 similar to *Tenupedunculus serrulatus* sp. n.); not evident if there is a rostrum or pseudorostrum; eyes reniform; appendix masculina not known.
- *Tenupedunculus drakensis* (Schultz, 1982). Tierra del Fuego, Argentina; 548 m; pseudorostrum rounded to acute; weak lateral spines on cephalon; reniform eyes; appendix masculina not known; male pereopod 1 not known. Originally placed in *Protallocoxa* Schultz, 1978 this species was later transferred to *Tenupedunculus* by Serov and Wilson (1995).
- *Tenupedunculus serraticaudum* (Kussakin & Vasina, 1984). South Atlantic; 500 m; pseudorostrum anteriorly round to acute; large lateral spines on cephalon; reniform eyes; appendix masculina blunt (excavate) with apical setae; male pereopod 1 carpus with superodistal process.

### Key to the species of Tenupedunculus sensu stricto

1	Lateral margins of the body (from cephalon to pleotelson) with serrations;
	shallow-water species, found at depths less than 50 m T. serrulatus sp. n.
_	Lateral margins of the body (from cephalon to pleotelson) without serrations;
	deep-water species, found at depths greater than 500 m2
2	Posterior margin of pleotelson distinctly produced T. virginale
_	Posterior margin of pleotelson not produced, obtusely or evenly rounded 3
3	Posterior margin of pleotelson obtusely rounded, with indications of uropo-
	dal bases
_	Posterior margin of pleotelson evenly rounded, without indications of uropo-
	dal bases

### Tenupedunculus serrulatus Song & Bruce, sp. n.

http://zoobank.org/EB46C41A-6548-4BBA-9C12-1B39042FF916 Figs 1–4

Material examined. All material from Capricorn Group, southern Great Barrier Reef. Holotype. ♂ (4.2 mm), 'Harry's Bommie, Heron Island, 23.46053°S, 151.9293°E, 13 November 2010, reef slope, dead *Acropora*, 9 m, CReefs stn. HI10-002C, coll. C. Buxton (MTQ W33638).

**Paratypes.** 2 (4.5, 4.1 mm [dissected]), same sample as holotype, (MTQ W52903).  $\bigcirc$  (5.8 mm [pereopod 1 dissected]), same data as holotype (MTQ W33654).  $\bigcirc$  (5.1 mm [pereopod 7 dissected]), Sykes Reef west, 23.4316°S, 152.0493°E, 14 November 2010, reef slope, 27 m, CReefs stn. HI10-009F, coll. J. Reimer (MTQ W33694). 5 ♀ (3.2-5.0 mm), 1 juv. (1.5 mm), Sykes Reef west, 23.4316°S, 152.0493°E, 14 November 2010, reef slope, 27 m, CReefs stn. HI10-009F, coll. J. Reimer (MTQ W33695). 20  $\Diamond$  and  $\heartsuit$ , same data as holotype, coll. C. Buxton, stn. HI10-002B (MTQ W33673, W33644) and HI10-002C (MTQ W33636, W33642). Q (3.5 mm), Lamont Reef, 23.5932°S, 152.0655°E, 16 November 2010, reef slope, dead Acropora, 9 m, CReefs stn. HI10-019B/1, coll. M. Blazewicz (MTQ W33753). 5 👌 (3.8–5.8 mm), Heron Island, southern side 'Twin Peaks', 23°28.357'S, 151°57.593'E, 28 November 2009, small rubble, 13-17 m CReefs stn. HI09-125F, coll. N.L. Bruce & K. Schnabel. (MTQ W52904). ♀ (5.8 mm), Heron Island 23.43238°S, 152.03375°E, 14 November 2009, CReefs stn. 018, no other data (MTQ W52905). ♂ (3.5 mm), 2 ♀ (2.8, 5.1 mm), Lamont Reef, southern side, 23°36.125'S, 152°03.152'E, 19 November 2009, coarse sand and small rubble, 9.7 m, CReefs stn. HI09-058D, coll. K. Schnabel & N.L. Bruce. (MTQ exW31591). MTQ W52906). 8  $\stackrel{?}{\rightarrow}$  and  $\stackrel{?}{\rightarrow}$ , Harry's Canyons, Heron Reef, 23°28.389'S, 151°57.835'E, 18 November 2009, reef slope, small rubble and coarse sand, 6 m, CReefs stn HI09-045D, coll. N.L. Bruce & K. Schnabel (MTQ W52907). 2 ♂, 2 ♀, 2 imm., Sykes Reef, 23°25.929'S, 152°02.924'E, 18 November 2009, 26 m, coll. S. Smith & A. Anderson (MTQ W52908).

**Non-type.** All Heron Island: north-eastern side, 20 November 2009, small rubble and sand at base of large bommies 7 m, CReefs stn HI09-064D (MTQ W31595). "The Patches" (=Mystery Bommie), 28 November 2009, rubble, mid-channel, 18 m, CReefs stn HI09-123C (MTQ W31604). "Harrys Bommie", 13 November 2010, dead coral on sandy bottom, CReefs stn HI10-002B, 10 m (MTQ W33669).

**Etymology.** From combining the Latin words '*serrula*' (serrated) and the ending of '*marginatus*', alluding to the serrated body margins of this species.

**Diagnosis (male).** Body (Fig. 1A) lateral margins with serrations. Pereonite 4 smallest. Pseudorostrum (Fig. 1C) wider than long, trapezoid-shape. Antennula (Fig. 1A, C) longer than cephalon, with ten flagellar articles. Antenna (Fig. 1A) longer than whole body length, with numerous flagellar articles. Maxilliped (Fig. 2D) endite distal margin with five fan setae. Pereopod 1 (Fig. 3A) superior carpal process distinctly long, bladelike. Uropods (Fig. 1A, H) well-developed, biramous, shorter than pleotelson; exopod shorter than endopod. Pleotelson (Fig. 4H) lateral margins with distinct notch.

**Description (male).** Body (Fig. 1A) length 3.3 times maximum width. Cephalon (Fig. 1C) length 0.5 times width, 1.6 times pereonite 1 length; lateral margins straight or very weakly convex, serrate, with two setae; antennal spines rounded; lateral spines moderate, acute and serrate, longer than antennal spines; space between lateral and antennal spines evenly rounded. Eyes (Fig. 1C) with seven ommatidia, pale brown, arranged in circle. Pereonites 1-7 (Fig. 1A) lateral margin serrate, with one seta. Sternal keel (Fig. 1B) present both in males and females as anteriorly directed spines on posteriorly directed spine on pereonites 6. Pereonite 1 length 0.3 times width, 0.9 times pereonite 2 length, width 1.1 times cephalon width. Pleotelson (Figs. 1A, 4H) length 0.9 times width; lateral margin serrate, with distinct notch.

*Antennula* (Fig. 1A, D) length 1.5 times cephalon length; article 1 length 1.6 times width, mesial margin with four short penicillate setae, distolateral margin with one large penicillate seta; article 2 length 1.8 times width, distomesial margin with one cluster of setae, including two penicillate seta, distolateral margin with one large penicillate seta and one short seta; article 3 length 2.9 times width, distomesial margin with one penicillate seta; flagellum with ten articles, one aesthetasc per article on distal nine articles.

Antenna (Fig. 1A, E, F, G) length approximately 1.5 times body length; peduncle article 1 length 0.8 times width, distolateral margin with one cluster of setae; article 2 length 0.6 times width, distal margin with two setae; article 3 length 1.1 times width, distomesial margin with two clusters of setae, mesial margin with one long seta and one short seta, lateral margin with five setae surrounding squama; article 4 length 0.7 times width, distomesial margin with three setae; article 5 length 6.8 times width; article 6 length 7.9 times width; each flagellum article with a cluster of four distally projecting setae, the cluster position serially repeating every four articles.

*Mandible* (Fig. 2A) left spine row with eleven spines, right spine row with six spines; palp article 1 length 2.5 times width, distolateral margin with one long seta, and two short setae; palp article 2 length 2.9 times width, with row of seven short serrate setae; article 3 length 2.6 times width. Maxillula (Fig. 2B) lateral lobe apex with 14 serrate RS; mesial



**Figure 1.** *Tenupedunculus serrulatus* sp. n., male holotype. **A** body, dorsal view **B** body, lateral view, sternal keel **C** cephalon, dorsal view **D** antennula **E** antenna **F** enlargement of peduncular articles 1–4 of antenna **G** enlargement of antennal flagellum articles **H** uropod. Scale bars: 1 mm (**A**, **B**), 0.5 mm (**C**), 0.2 mm (**D**, **F**, **G**, **H**), 0.4 mm (**E**).



**Figure 2.** *Tenupedunculus serrulatus* sp. n., male holotype. **A** mandible with palp **B** maxillula, with details of mesial and lateral lobes **C** maxilla **D** maxilliped, with enlargement of endite. Scale bar: 0.1 mm.

lobe apex with two large plumose setae, distomesial margin with one setulate RS, one large plumose seta. Maxilla (Fig. 2C) mesial lobe mesial margin with eight large plumose setae, apex with three large setulate setae, two setae with spatulate tips; middle lobe apex with four large setulate setae; lateral lobe apex with four large setulate setae. Maxilliped (Fig. 2D) basis length 2.2 times maximum width, width 1.1 times endite width; endite distal margin with five fan setae, distomesial margin with six serrate setae, distomesial corner with two triangular RS; epipod length 3.2 times width, width 1.2 times basis width, apex acute, distomesial margin with eight regularly spaced setae, lateral margin sinuate.

*Pereopod 1* (Fig. 3A) basis length 3.7 times width; superior margin with three long setae alternate with three short; submarginal row of short setae.

*Pereopod 1* ischium length 1.6 times width; inferior margin with one short seta; superodistal margin produced, with eight long setae, apex acute.

*Pereopod 1* merus angular; merus length 1.1 times width, 0.8 times carpus length, 0.8 times ischium length; inferior margin with one short seta, two long setae; inferior submargin with a dense patch of long setae; distal margin with no setae; superodistal margin produced, apex acute, densely setose with long setae and two short setae.

*Pereopod 1* carpus triangular; length 1.5 times width, 1.1 times ischium length; distal margin convex, with two short setae; inferior margin clearly defined, and densely setose along distal two-thirds only; inferior submargin with a dense patch of short setae. Superior carpal process long, bladelike; length 3.3 times width, 1.7 times carpal width; extending distally approximately half length of propodus; apex acute, densely setose; inferior margin smooth, straight, densely setose along distal two-thirds only, with several rows of setae; superior margin slightly convex, setose along full length.

*Pereopod 1* propodus robust superiorly with inferior side of article flattened; length 1.5 times maximum width, 3.6 times proximal width, 2.2 times ischium length; inferior margin clearly defined, long, 0.7 times propodus length, 0.6 times superior margin length, densely covered with rows of long and serrate setae and with submarginal row of short setae; superior margin setose, setae regularly spaced. Propodal palm (Fig. 3B) width 0.6 times maximum propodus width, slightly oblique; toothed lobe with four teeth, largest tooth length 3.0 times smallest tooth length; short setae inserting between teeth, cluster of setae at articulation and long setae on mesial surface.

*Pereopod 1* dactylus robust; length 4.8 times width, 1.4 times propodal palm width, 0.9 times propodus distal width (not including process), 0.6 times propodus length; superior margin distal third setose, with regularly spaced setae; distal margin setae regularly spaced along entire length; and more sparsely distributed long setae; mesial surface sparsely setose.

*Pereopod 2* (Fig. 3C) basis superior margin with three penicillate setae; ischium superior margin with one large seta; merus superodistal margin produced with one large RS at apex; carpus superodistal margin with cluster of setae, including one penicillate seta, inferior margin with four flagellated RS (most distal paired with one RS); propodus superodistal margin with cluster of setae, including one penicillate seta, inferior margin with cluster of setae, including one penicillate seta, inferior margin with cluster of setae, including one penicillate seta, inferior margin with four flagellated RS, inferodistal margin with one flagellated RS.



**Figure 3.** *Tenupedunculus serrulatus* sp. n., **A–D** male holotype **E**, **F** female paratype. **A** pereopod 1 **B** enlargement of pereopod 1 palm and dactylus **C** pereopod 2 **D** pereopod 7 **E** pereopod 1 **F** enlargement of pereopod 1 palm and dactylus. Scale bars: 0.2 mm (**A**, **C**, **D**, **E**), 0.1 mm (**B**, **F**).

*Pereopod 7* (Fig. 3D) basis superior margin with two penicillate setae; inferior margin without stiff setae; carpus inferior margin with two flagellated RS; propodus inferior margin with five flagellated RS, inferodistal margin with one flagellated RS.

*Pleopod 1* (Fig. 4A) protopod length 0.7 times width, distal margin with pair of robust setae, surface setae absent; rami lateral margins with regularly spaced setae along distal two-thirds of margin, inferior surface without setae. *Pleopod 2* (Fig. 4B) protopod length 2.6 times medial width, basal lobe width 1.8 times medial width, distal lobe distinctly shorter than exopod, distal lobe blunt; endopod length 0.6 times protopod length, without setae; *appendix masculina* (Fig. 4C) length 1.6 times endopod length, 0.9 times protopod length, widest distally; lateral margin without distal groove; mesial margin without setae; apex convex, depression fringed with scale setae; lateral margin without setae. *Pleopod 3* (Fig. 4D) endopod apex with five plumose setae. *Pleopod 4* (Fig. 4E) exopod apex with nine plumose setae. *Pleopod 5* (Fig. 4F) apex with five plumose setae.

*Uropod* (Fig. 1A, H) length 0.2 times body length, 0.7 times pleotelson length; protopod length 2.4 times width; endopod length 1.1 times protopod length, distal and sub-distal margins with three penicillate setae, distal tip with cluster of elongate setae with maximum length 1.5 times endopod length; exopod length 0.8 times protopod length, 0.7 times endopod length, distal tip with cluster of elongate setae with maximum length 0.9 times exopod length.

**Description (female).** *Pereopod 1* (Fig. 3E) basis length 3.2 times width, superior margin with 12 short setae, inferior margin with six short setae. Ischium superodistal margin produced, apex acute. Merus superodistal margin produced; process apex acute. Propodus length 1.4 times distal (maximum) width, 2.8 times proximal width, 1.6 times ischium length; inferior margin length 0.7 times propodus length, densely setose, with a row of setulate setae and a row of simple setae, setae restricted to distal three-quarters of margin; propodal palm (Fig. 3F) width 0.8 times distal propodus width; straight, slightly oblique, with five teeth, inferior end with single robust flagellated seta, and single robust serrate seta adjacent to first tooth, articulation with 14 long setae. Dactylus length 5.6 times width, 1.3 times palm width, inferior margin with 17 regularly spaced robust flagellated setae, with irregularly distributed long setae.

*Pleopod 2* (Fig. 4G) length 1.5 times maximum width; lateral margins with 2–5 setae; not parallel, narrowing posteriorly; posterolateral margins concave, apex slightly notched, asymmetric, with two setae; inferior surface without setae.

**Size.** Males 2.6–5.1 mm (mean 3.9 mm, n = 6); females 2.3–5.8 mm (mean 3.8 mm, n = 24).

**Remarks.** *Tenupedunculus serrulatus* sp. n. is distinguished from other species of *Tenupedunculus* by the following unique combination characters: merus superodistal margin with strongly produced distally rounded process, more than 3.0 times as long as wide; male pleopod 2 appendix masculina bluntly rounded apically, without apical setae; and the lateral margins of the body (from cephalon to pleotelson) are serrated.

*Tenupedunculus serrulatus* is most similar to the deep-water species *T. pulchrum* (Schultz, 1982) with regard to general external appearance, but is distinguished by the



**Figure 4.** *Tenupedunculus serrulatus* sp. n., **A–F, H** male holotype **G** female paratype. **A** pleopod 1 **B** pleopod 2 **C** enlargement of appendix masculina apex **D** pleopod 3 **E** pleopod 4 **F** pleopod 5 **G** pleopod 2 **H** pleotelson, dorsal view. Scale bars: 0.1 mm (**A**, **G**), 0.05 mm (**B**, **C**), 0.2 mm (**D–F**), 0.5 mm (**H**).

following: small body size (< 6 mm vs. 9 mm for the holotype of *T. pulchrum*), serrations on lateral margin of the whole body (vs. smooth); the very strongly produced process on pereopod 1 carpus superodistal margin, approximately 2.5 times as long as width of carpus proximal margin (vs. weak process, approximately 0.6 times as long as width of carpus proximal margin); and appendix masculina without any acute part (vs. with small acute part). *Tenupedunculus serrulatus* is the only shallow-water tropical species in the genus.

**Distribution.** Heron Island and reefs of the Capricorn Group, southern Great Barrier Reef, Australia; at depths of 6–27 m.

# Genus Stenobermuda Schultz, 1979

Stenobermuda Schultz, 1979: 905.– Kensley and Schotte 1989: 106.– Serov and Wilson 1995: 77.– Kensley and Schotte 2002: 1456.

Stenetrigus Schultz, 1982: 58.

**Type species.** *Stenobermuda acutirostrata* Schultz, 1979; by original designation and monotypy.

**Species included.** *Stenobermuda acutirostrata* Schultz, 1979, Bermuda (type species); *S. brucei* Kensley & Schotte, 2002, Zanzibar, Tanzania; *S. iliffei* Kensley, 1994, Bermuda; *S. mergens* Botosaneanu & Iliffe, 1999, Bahamas; *S. syzygus* (Barnard, 1940), South Africa.

**Remarks.** *Stenobermuda* is a small genus with both marine and stygobiont species. Recent diagnoses do not require modification in light of other recent revisions, and the genus can be readily recognised by the prominent, narrow, and acute rostral process, small or absent eyes, and pereopod 1 articles without prominent processes or an expanded propodus. Sexual dimorphism is weak in the genus.

A diagnostic character of the genus *Stenobermuda* Schultz, 1979 is the acute and distinct rostrum, but one species, *S. iliffei* Kensley, 1994 is described as having a rostrum but figured with a pseudorostrum. The presence or absence of a rostrum of *S. iliffei* therefore does need to be confirmed to assess its status within the genus. Other than the apparent difference is rostrum the species agrees entirely with *Stenobermuda*.

#### Key to the species of Stenobermuda

Cave species are indicated by brackets.

1	Eyes (ommatidia) absent	[S. mergens]
_	Eyes (ommatidia) present	2
2	Dorsal coxal plates absent	
_	Dorsal coxal plates present	

3	Body length < 3 mm; pereopod 1 propodus narrow, length 1.9 times maxi-
	mum width
_	Body length > 6 mm; pereopod 1 propodus expanded, length 1.1 times maxi-
	mum width
4	Eyes with five ommatidia; dorsal coxal plates large
_	Eyes with four ommatidia; dorsal coxal plates small5
5	Pereopod 1 propodus expanded, length 1.1 times maximum width; rostrum
	proximal lateral margin straightS. brucei
_	Pereopod 1 propodus narrow, length 1.9 times maximum width; rostrum
	proximal lateral margin convex

### Stenobermuda warooga Song & Bruce, sp. n.

http://zoobank.org/720698E6-C575-4253-97E3-4EFE0D82F482 Figs 5–8

**Material examined. Holotype.** ♂ (1.6 mm), Yonge Reef, northern Great Barrier Reef, 14.57302°S, 145.6189°E, 10 September 2010, outer reef slope, coarse sand, 25 m, CReefs stn. L110-126B (MTQ W32968).

**Paratypes.** 3 ♂ (1.7 [all appendage dissected], 1.2 [antennula and pleopod 2 dissected], 1.5 mm [pleopod 1 dissected]), same data as holotype (MTQ W52909). 3 ♂ (1.4, 1.5 [dissected], 1.7 mm), High Rock (between Direction Islands and Ribbon Reef No. 10), northern Great Barrier Reef, 14.82462°S,145.552°E, 6 September 2010, coral rubble, 8 m, CReefs stn. LI10-092A, coll. C. Buxton (MTQ W32917). ♂ (1.2 mm), 14.57302°S, 145.61980°E, Yonge Reef, 10 November 2010, outside; small coral rubble in spur, 20 m CReefs stn LI10-126A (MTQ W52910). ♂ (1.4 mm), Yonge Reef, 14.60681°S, 145.6311°E, 20 February 2009, outer reef front., dead coral, 30 m, coll. CReefs stn LI09-15B Shawn Smith & Julian Caley (MTQ W52911).

**Etymology.** The epithet '*warooga*' is an Aboriginal word meaning small child, in reference to the small size of this species; noun in apposition.

**Diagnosis (male).** Body (Fig. 5A) lateral margins smooth. Pereonite 4 smallest. Rostrum (Fig. 5B) acute, proximal lateral margin convex. Antennula (Fig. 5C) shorter than cephalon, with three flagellar articles. Antenna (Fig. 5D) shorter than whole body length, with numerous flagellar articles. Maxilliped (Fig. 6D) endite distal margin with three fan setae. Pereopod 1 (Fig. 7A) superodistal and inferodistal margin without process. Uropod (Fig. 5A, G) very short, biramous; exopod shorter than endopod. Pleotelson (Fig. 8G) lateral margins with distinct notch.

**Description (male).** *Body* (Fig. 5A) length 3.6 times maximum width. *Cephalon* (Fig. 5B) length 0.8 times width, 3.2 times pereonite 1 length; lateral margins straight or very weakly convex, smooth, with one setae; antennal spines acute; lateral spines acute, longer than antennal spines. *Rostrum* (Fig. 5B) proximal lateral margin convex. *Eyes* (Fig. 5B) with four ommatidia, pale brown, arranged in circle. *Pereonites* 1–7



**Figure 5.** *Stenobermuda warooga* sp. n., male holotype. **A** body, dorsal view **B** cephalon, dorsal view **C** antennula **D** antenna **E** enlargement of peduncular articles 1–4 of antenna **F** enlargement of antennal flagellum articles **G** uropod. Scale bars: 0.25 mm (**A**), 0.125 mm (**B**), 0.05 mm (**C**, **E**, **F**, **G**), 0.2 mm (**D**).



**Figure 6.** *Stenobermuda warooga* sp. n., male holotype. **A** mandible with palp **B** maxillula, with details of mesial and lateral lobes **C** maxilla **D** maxilliped, with enlargement of endite. Scale bars: 0.05 mm.



**Figure 7.** *Stenobermuda warooga* sp. n., male holotype. **A** pereopod 1 **B** enlargement of pereopod 1 palm and dactylus **C** pereopod 2, with enlargement of dactylus **D** pereopod 7, with enlargement of dactylus. Scale bars: 0.05 mm.



**Figure 8.** *Stenobermuda warooga* sp. n., male holotype. **A** pleopod 1 **B** pleopod 2 **C** enlargement of appendix masculina apex **D** pleopod 3 **E** pleopod 4 **F** pleopod 5 **G** pleotelson, dorsal view. Scale bars: 0.05 mm (**A**, **B**), 0.025 mm (**C**), 0.1 mm (**D**–**F**), 0.25 mm (**G**).

(Fig. 5A) lateral margin smooth, without setae; pereonite 1 length 0.2 times width, 0.8 times pereonite 2 length, width 1.1 times cephalon width; pereonite 5–7 distolateral margin not produced. *Coxal plates* (Fig. 5A) small, visible dorsally on pereonites 4–6. *Pleotelson* (Figs 5A, 8G) length 0.9 times width, with distinct notch.

*Antennula* (Fig. 5C) length 0.7 times cephalon length; article 1 length 1.6 times width, mesial margin with one short penicillate setae, distolateral margin with one large penicillate seta; article 2 length 2.3 times width, distolateral margin with one large penicillate seta; article 4 length 0.5 times width; flagellum with three articles, one aesthetasc per article on distal two articles.

Antenna (Fig. 5D, E, F) length approximately 0.7 times body length; peduncle article 1 length 0.8 times width; article 2 length 0.9 times width, distolateral margin with one long seta; article 3 length 1.3 times width, distomesial margin with one cluster of setae, lateral margin with six setae surrounding squama; each flagellum article with a cluster of four distally projecting setae, the cluster position serially repeating every four articles.

*Mandible* (Fig. 6A) left spine row with four spines, right spine row with six spines; palp article 1 length 3.5 times width, distolateral margin with one long seta; palp article 2 length 2.7 times width, with row of one short serrate setae; article 3 length 2.8 times width. *Maxillula* (Fig. 6B) lateral lobe apex with seven serrate RS; mesial lobe apex with three large plumose setae. *Maxilla* (Fig. 6C) mesial lobe mesial margin with four large plumose setae; lateral lobe apex with four large setulate setae; middle lobe apex with three large setulate setae; lateral lobe apex with four large setulate setae. *Maxilliped* (Fig. 6D) basis length 2.0 times maximum width, width 0.9 times endite width; endite distal margin with three fan setae, distomesial corner with one triangular RS; epipod length 3.4 times width, width 1.1 times basis width, apex acute, lateral margin evenly convex.

*Pereopod 1* (Fig. 7A) basis length 3.2 times width; superior margin with three short setae; submarginal row of short setae.

*Pereopod 1* ischium length 1.9 times width; inferior margin with one short seta; distal margin with one short setae; superodistal margin not produced, apex rounded.

*Pereopod 1* merus rectangular; merus length 1.4 times width, 0.8 times carpus length, 0.9 times ischium length; inferior margin with two short setae, one long seta; distal margin with no setae; superodistal margin not produced, apex rounded, with two short setae.

*Pereopod 1* carpus rectangular; length 2.0 times width, 1.3 times ischium length; distal margin convex; inferior margin clearly defined, and with four stout setae and four long setae; superodistal margin not produced, apex obliquely truncate.

*Pereopod 1* propodus robust and narrow; length 1.9 times maximum width, 3.2 times proximal width, 1.3 times ischium length; inferior margin clearly defined, long, 0.7 times propodus length, 0.7 times superior margin length, lightly setose, regularly spaced setae along entire length; superior margin setae absent. Propodal palm (Fig. 7B) width 0.7 times maximum propodus width, slightly oblique; with three large serrate setae and nine long setae. Pereopod 1 dactylus narrow.

*Pereopod 1* dactylus convex in mid-section; length 1.9 times width, 1.3 times propodal palm width, 0.8 times propodus distal width (not including process), 0.4 times

	Total length (mm) of adult male	Rostrum (Pseudorostrum), proximal lateral margin	Rostrum/ Pseudorostrum	Eyes (ommatidia)	Dorsally visible coxae (size)	Pereopod 1 propodus length
<i>S. warooga</i> sp. n.	1.6	Convex	Rostrum	Small rounded (4 ommatidia)	Pereonites 4–6 (small)	Narrow, 1.9 times maximum width
S. acutirostrata	4.8	Straight	Rostrum	Small rounded (5 ommatidia)	Pereonites 1, 4–6 (large)	Normal, 1.6 times maximum width
S. brucei	3.1	Straight	Rostrum	Small rounded (4 ommatidia)	Pereonites 4–6 (small)	Expanded, 1.1 times maximum width
S. iliffei	2.9	Straight	Pseudorostrum (?)	Small rounded (4 ommatidia)	Not visible	Narrow, 1.9 times maximum width
S. mergens	3.2	Straight	Rostrum	Absent	Pereonites 1–6 (small)	Normal, 1.6 times maximum width
S. syzygus	6.5	Straight	Rostrum	Small rounded (4 ommatidia)	Not visible	Expanded, 1.1 times maximum width

**Table 2.** Comparison of diagnostic characters between *S. warooga* sp. n. and other species of *Stenober-muda* (male).

propodus length; superior margin distal third with four long setae. Distal margin with three serrate setae. Mesial surface not setose.

*Pereopod 2* (Fig. 7C) basis medial inferior margin with stiff seta absent; ischium superior margin with stiff setae absent; merus superodistal margin RS absent; carpus superodistal margin with three setae, inferior margin with two flagellated RS (most distal paired with one RS); propodus superodistal margin with three setae, inferior margin wi

*Pereopod 7* (Fig. 7D) basis inferior margin without stiff setae; carpus inferior margin with two flagellated RS; propodus inferior margin with one flagellated RS, inferodistal margin with one flagellated RS.

*Pleopod 1* (Fig. 8A) protopod length 0.7 times width, surface setae present; rami lateral margins with regularly spaced setae along distal half, inferior surface without setae. *Pleopod 2* (Fig. 8B) protopod length 1.9 times medial width, basal lobe width 1.4 times medial width, distal lobe distinctly shorter than exopod, distal lobe blunt; endopod length 0.7 times protopod length, without setae; *appendix masculina* (Fig. 8C) length 1.8 times endopod length, 1.3 times protopod length, widest distally; lateral margin without distal groove; mesial margin without setae; apex with two process and cuticular fan; lateral margin without setae. *Pleopod 3* (Fig. 8D) endopod apex with three plumose setae. *Pleopod 4* (Fig. 8E) exopod apex with four plumose setae. *Pleopod 5* (Fig. 8F) apex with one seta.

*Uropod* (Fig. 5A, G) very short, length 0.05 times body length, 0.2 times pleotelson length; protopod length 1.4 times width; endopod length 1.5 times protopod length, distal and sub-distal margins with one penicillate setae, distal tip with cluster of elongate setae with maximum length 1.9 times endopod length; exopod length 0.9 times protopod length, 0.6 times endopod length, distal tip with cluster of elongate setae with maximum length 3.6 times exopod length. Female. Not known.

**Size.** Males 1.2–1.7 mm (mean 1.5 mm, *n* = 9).

**Remarks.** *Stenobermuda warooga* sp. n. can be identified by the following unique combination characters: small body size of the adult male (< 2 mm); rostrum proximal lateral margin with convex margin; pereonite 5 distolateral margin not produced; pereopod 1 propodus narrow, length 1.9 times maximum width (Table 2). The most similar species is *S. brucei* Kensley & Schotte, 2002, a species also occurring on coral reefs, with regard to external appearance. However, propodus of pereopod 1 is strongly expanded, length 1.1 times maximum width, and proximal lateral margin of rostrum is straight in *S. brucei* (propodus of pereopod 1 is narrow, length 1.9 times maximum width, and proximal lateral margin of rostrum is convex in *S. warooga* sp. n.).

**Distribution.** Yonge Reef and High Rock, Lizard Island region, northern Great Barrier Reef both outer barrier reefs; at depths of 8–30 m.

### Acknowledgements

This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR201524202, NIBR201624202). J.-H. Song thanks the Museum of Tropical Queensland, Queensland Museum for provision of facilities during his visit.

The authors gratefully acknowledge Australian Biological Resources Study for providing support to NLB for this study (ABRS grant 2009-30). Material was collected under the auspices of the CReefs project organised by the Australian Institute of Marine Science (AIMS), Townsville. The CReefs Australia Project was generously sponsored by BHP Billiton in partnership with The Great Barrier Reef Foundation, the Australian Institute of Marine Science and the Alfred P. Sloan Foundation; CReefs is a field program of the Census of Marine Life. The authors thank Julian Caley and Shawn Smith (AIMS) for their excellent organisation and field support; Magda Błażewicz-Paszkowycz (University of Lodz, Poland) and Kareen Schnabel (NIWA, Wellington, New Zealand), Slava Ivanenko (Moscow State University, Russia), and James Reimer (University of the Ryukyus, Japan) for their excellent help while collecting at Lizard and Heron Islands. This is contribution number 204 of the NWU Water Research Group.

### References

- Barnard KH (1940) Contribution to the crustacean fauna of South Africa. XII. Further additions to the Tanaidacea, Isopoda, and Amphipoda, together with keys for the identification of hitherto recorded marine and freshwater species. Annals of the South African Museum 32: 381–543.
- Beddard FE (1886) Preliminary notice of the Isopoda collected during the voyage of H.M.S. *Challenger.* Part III. Proceedings of the Zoological Society of London 1886(7): 97–122.

- Botosaneanu L, Iliffe TM (1999) A simple scenario for stygobitization in *Stenobermuda* Schultz, 1978 (Isopoda Asellota Stenetriidae), with description of a new species from Andros Island, Bahamas. Journal of Speleology 26: 37–45. https://doi.org/10.5038/1827-806X.26.1.4
- Bruce NL (2015) Joeropsididae Nordenstam, 1933 (Crustacea: Isopoda: Asellota) from the Lizard Island region of the Great Barrier Reef, Queensland, Australia. ZooKeys 491: 1–62. https://doi.org/10.3897/zookeys.491.4932
- Bruce NL, Buxton CL (2013) Review of the marine isopod crustacean genus *Hansenium* Serov & Wilson, 1995 (Asellota: Stenetriidae) from tropical Australia and Papua New Guinea, with description of a new genus. Zootaxa 3664(4): 445–478. https://doi.org/10.11646/ zootaxa.3664.4.3
- Bruce NL, Cumming RL (2015) A new genus of Stenetriidae (Asellota: Isopoda: Crustacea) from the Great Barrier Reef, Australia and the southwestern Pacific. Zootaxa 3941(4): 485–508. https://doi.org/10.11646/zootaxa.3941.4.2
- Coleman CO, Lowry JK, Macfarlane T (2010) DELTA for beginners. An introduction into the taxonomy software package DELTA. ZooKeys 45: 1–75. https://doi.org/10.3897/zookeys.45.263
- Dallwitz MJ, Paine TA, Zurcher EJ (2006) User's guide to the DELTA system: A general system for processing taxonomic descriptions. http://delta-intkey.com/ [accessed March 2016]
- Hansen HJ (1905) On the morphology and classification of the Asellota group of crustaceans with descriptions of the genus *Stenetrium* Haswell and its species. Proceedings of the Zoological Society of London 1904 (2 Suppl. II): 302–331.
- Haswell WA (1881) On some new Australian marine Isopoda. Part I. Proceedings of the Linnean Society of New South Wales 5(4): 470–481, pls 16–19.
- Kensley B (1984a) The Atlantic barrier reef ecosystem at Carrie Bow Cay, Belize, III: new marine Isopoda. Smithsonian Contributions to the Marine Sciences 24: 1–81. https://doi. org/10.5479/si.01960768.24.1
- Kensley B (1984b) The role of isopod crustaceans in the reef crest community at Carrie Bow Cay, Belize. Marine Ecology 5(1): 29–44. https://doi.org/10.1111/j.1439-0485.1984. tb00305.x
- Kensley B (1988) Preliminary observation on the isopod crustacean fauna of Aldabra Atoll. Bulletin of the Biological Society of Washington 8: 40–44.
- Kensley B (1994) Records of shallow-water marine isopods from Bermuda with descriptions of four new species. Journal of Crustacean Biology 14: 319–336. https://doi. org/10.2307/1548912
- Kensley B, Schotte M (1989) Guide to the Marine Isopod Crustaceans of the Caribbean. Smithsonian Institution Press, Washington, D.C. & London, 308 pp.
- Kensley B, Schotte M (2002) New species and records of Asellota from the Indian Ocean (Crustacea: Peracarida: Isopoda). Journal of Natural History 36(12): 1421–1461. https:// doi.org/10.1080/00222930110050401
- Kussakin OG (1967) Fauna of Isopoda and Tanaidacea in the coastal zones of the Antarctic and subantarctic water. In: Andriyashev AP, Ushakov PV (Eds) Biological Reports of the Soviet Antarctic Expedition (1955–1958). Akademii Nauk SSSR, Leningrad, 220–380.
- Kussakin OG, Vasina GS (1984) Deep-sea lower asellotes from the Scotia Sea and South Sandwich Trench. Biologiya Morya, Vladivostok, 6: 9–17.

- Martin JW, Heard RW, Wetzer R (2003) A new species of *Stenetrium* Haswell, 1881 (Crustacea: Peracarida: Asellota), from Navassa Island, northern Caribbean Sea. Proceedings of the Biological Society of Washington 116(4): 967–977.
- Müller H-G (1990) Stenetriidae from the Caribbean Sea of N-Columbia. (Crustacea: Isopoda: Asellota). Senckenbergiana Biologia 70(4/6): 397–404.
- Müller H-G (1991a) The marine isopod family Stenetriidae from the coral reefs at Bora Bora and Moorea, Society Islands, with descriptions of four new species (Crustacea). Revue suisse de Zoologie 98(1): 51–76. https://doi.org/10.5962/bhl.part.79777
- Müller H-G (1991b) Stenetriidae from coral reefs at Réunion Island, southern Indian Ocean. Description of three new species (Crustacea: Isopoda: Asellota). Senckenbergiana Biologia 71(4/6): 303–318.
- Schultz GA (1978) Protallocoxoidea new superfamily (Isopoda Asellota) with a description of *Protallocoxa weddellensis* new genus, new species from the Antarctic Ocean. Crustaceana 34(3): 245–250. https://doi.org/10.1163/156854078X00808
- Schultz GA (1979) A new Asellota (Stenetriidae) and two, one new, Anthuridea (Anthuridae) from Bermuda (Crustacea, Isopoda). Proceedings of the Biological Society of Washington 91(4): 904–911 [for 1978].
- Schultz GA (1982) Species of Protallocoxoidea and Stenetrioidea (Isopoda, Asellota) from Antarctic and southern seas. Biology of the Antarctic Seas 32: 17–62.
- Serov PA, Wilson GDF (1995) A review of the Stenetriidae (Crustacea: Isopoda: Asellota). Records of the Australian Museum 47: 39–82. https://doi.org/10.3853/j.0067-1975.47.1995.2
- Vanhöffen E (1914) Die Isopoden der Deutschen Südpolar Expedition 1901–1903. Deutschen Südpolar Expedition 7(4): 447–598.
- Vasina GS (1982) A new species of the genus *Stenetrium* (Crustacea, Isopoda, Stenetriidae) from macrophytic of the Patagonian Shelf. Fauna and Distribution of Crustaceans from the Southern and Antarctic Waters. Akademiya Nauk CCCP, Dal'nevostochnyi Nauchyi Tsentr (Far East Science Center), Vladivostok, 106–109.
- Wilson GDF, Schotte M (2008) Stenetriidae Hansen, 1905. In: Boyko CB,Bruce NL, Hadfield KA, Merrin KL, Ota Y, Poore GCB, Taiti S, Schotte M, Wilson GDF (Eds) (2008 onwards) World Marine, Freshwater and Terrestrial Isopod Crustaceans database. World Register of Marine Species at http://www.marinespecies.org/aphia.php?p=taxdetails&id=118270 [accessed 2017-11-23]

RESEARCH ARTICLE



# Ampithoidae (Crustacea, Amphipoda) from New Zealand

Rachael A. Peart<sup>1</sup>, Anne-Nina Lörz<sup>2</sup>

**1** P.O. Box 14901, Kilbirnie, Wellington, 6241, New Zealand **2** CeNak, Zoologisches Institut und Museum, Martin-Luther-King Platz 3, 20146 Hamburg, Germany

Corresponding author: Rachael A. Peart (rachael.peart@niwa.co.nz)

Academic editor: S. Brix   1	Received 12 June 2017   Accepted 11 September 2017   Published 26 January 2018
h	tp://zoobank.org/121ED460-2B81-4911-B147-A3D6AB04652D

**Citation:** Peart RA, Lörz A-N (2017) Ampithoidae (Crustacea, Amphipoda) from New Zealand. ZooKeys 733: 25–48. https://doi.org/10.3897/zookeys.733.14052

### Abstract

Ampithoidae is a family of marine Amphipoda with approximately 230 species, belonging to 16 genera. The family has a worldwide distribution as algal dwellers. So far only five species are known from New Zealand. Recent collections and examination of historic collection material added two new species, which are described herein. An overview of and a key to the New Zealand Ampithoidae is provided.

### **Keywords**

Algae dweller, new species, Peracarida, New Zealand

# Introduction

The family, Ampithoidae, is a broadly distributed group of primarily algal-dwelling amphipods. They have been well described and reported from North and South American-, European-, African-, Australian- and Asian waters. The Ampithoidae record is quite sparse in boreal waters, but not unknown (De Broyer et al. 2007). Globally there are 231 species (Horton et al. 2017). However, despite this diversity, there have only been five species recorded from New Zealand.

The major source of information on New Zealand ampithoid amphipods is J.L. Barnard's detailed monograph of algal dwelling gammarid amphipods (1972). This

Copyright Rachael A. Peart, Anne-Nina Lörz. This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

paper documents four species of this family all in the genus, *Ampithoe* Leach, 1814. One of these species was originally described by Hurley (1954) (as *Pleonexes lessoniae* Hurley, 1954) and was subsequently designated the type species for the genus *Pseu-dopleonexes* (Conlan, 1982), two others were new descriptions (*Ampithoe hinatore* and *Ampithoe aorangi*) and the last an undesignated species (*Ampithoe* sp.). Recent work (Hughes and Peart 2015, Peart 2017, and this current work) has expanded and revised this previous research.

Using freshly collected material and type material from the NIWA Invertebrate collection, this paper provides a checklist to the Ampithoids from New Zealand waters, providing a diagnosis for each species and a key to the species of the family. Adding to the count of species from these is the description of two new species, *Exampithoe plumosa* sp. n. and *Pseudopleonexes evensis* sp. n.

# Materials and methods

Recent collections were sampled via snorkelling from *Macrocystis* sp. in Wellington and collected from a large, drifting detached plant of *Durvillaea antarctica* from Otago Harbour. They were immediately preserved in 95% ethanol. Specimens were examined and dissected using a Leica MZ12.5, in Wellington and drawn using a camera lucida attachment. Small appendages (mouthparts, uropods, telson) were temporarily mounted in glycerin and examined and drawn using a compound microscope (Zeiss, in Wellington) fitted with a camera lucida. The body lengths of specimens examined were measured by tracing individual's mid-trunk lengths (tip of the rostrum to end of telson) using a camera lucida. For scanning electron microscope (SEM) imaging the specimens and appendages were dehydrated through a graduated ethanol series, acetone dried, mounted on studs, coated with gold-palladium and investigated via a SEM LEO1525.

Type material and other material examined is held at the National Institute of Water and Atmospheric Research Invertebrate Collection at Wellington, New Zealand (NIWA) and the CeNak, Zoological Museum Hamburg.

# **Systematics**

# Order AMPHIPODA Latreille, 1816 Suborder SENTICAUDATA Lowry & Myers, 2013

### Ampithoidae Boeck, 1870

In New Zealand waters, the fauna of the family Ampithoidae is represented by four genera comprised of seven species, two of which are newly described here.

### Ampithoe Leach, 1814

# Ampithoe hinatore J.L. Barnard, 1972

Ampithoe hinatore J.L. Barnard, 1972: 39-42, figs 11-12.

**Type material.** Holotype: male, 9.0 mm, NIWA 831, station E970, Kaikoura, New Zealand, 42.417°S 173.700°E, intertidal wash of algae, 22 January, 1968.

Paratype: Female, 7.3 mm, 2 specimens, NIWA 832, station E970, Kaikoura, New Zealand, 42.417°S 173.700°E, intertidal wash of algae, 22 January, 1968.

Diagnosis. Male: Eye prominent. Epistome and upper lip, in situ, directed straight down, perpendicular to the head. Lower lip outer plate notched, outer lobe extending past inner lobe. Mandible molar well developed and triturating, palp robust and three-articulate, article three distally rounded. Maxilla 1 palp well developed. Pereopods weakly setose. Gnathopod 1 weakly sexually dimorphic; coxa produced anteriorly; basis anteroventral lobe prominent; propodus subovoid, anterodistal setose lobe absent, palm acute and concave defined by a subacute posterodistal tooth and robust seta; dactylus subequal in length to palm. Gnathopod 2 more robust and slightly longer than gnathopod 1; basis anteroventral lobe large and setose; carpus subtriangular; propodus longer than carpus, propodus broad, anterodistal lobe absent, palm acute, defined by a subacute posterodistal tooth and robust seta; dactylus subequal in length to palm. Pereopods 3 and 4 similar in size and shape; basis slightly expanded and glandular; merus narrow, lobe absent. Pereopod 5 basis rounded; distal articles slender; propodus weakly prehensile. Pereopods 6 and 7 similar, increasing in length; distal articles slender; propodus weakly prehensile. Epimeron 3 posteroventral corner rounded without tooth. Uropod 1, in situ, reaching to the end of uropod 2, peduncle distoventral spur absent. Uropod 2 peduncle rounded lateral distoventral process absent. Uropod 3 broad, peduncle with six distal robust setae; rami short; outer ramus with two recurved robust setae, denticle patch; inner ramus with robust and slender distal setae. Telson subtriangular with reduced, small cusps, denticles absent, with lateral and apical setae.

Female. Similar to male, except: Gnathopods 1-2 palms less excavate.

**Remarks.** Known only from the type locality, Kaikoura on the New Zealand, South Island north-eastern coast. J.L. Barnard (1972) recorded only three specimens, whilst Fenwick (1976) also noted the presence of this species in a wave exposure study, but in relatively small abundances (16 out of around 60,000 individuals). Barnard (1972) notes that it has a similar morphology to *Ampithoe waialua* Barnard, 1970 from Hawaii, which is also similar to the *Ampithoe ramondi* Audouin, 1826 complex of species. Lowry (1974) refers to the presence of *Ampithoe hinatore* from Kaikoura, however, these are not new records just repeats from Barnard's paper.

Distribution. Kaikoura, South Island, New Zealand

# Exampithoe K.H. Barnard, 1925

### Exampithoe plumosa sp. n.

http://zoobank.org/9ABFEAC2-509F-42C0-936A-C3FAABB215B2 Figs 1–4

**Type material.** Holotype, male, 10 mm, NIWA 121270, KH-NZ1-9, from drifting *Durvillaea antarctica* raft from near Taiaroa Head, inside Otago Harbour, Dunedin, New Zealand, 45°46'19"S, 170°43'30"E, 0 m depth, 22 January 2010, J. Waters.

Paratypes: Female, 7 mm, NIWA 121269, Male, 9 mm, ZMH K-46915, KH-NZ1-9, from drifting *Durvillaea antarctica* raft near Taiaroa Head, inside Otago Harbour, Dunedin, New Zealand, 45°46'19"S, 170°43'30"E, 0 m depth, 22 January 2010, J. Waters.

**Diagnosis.** Male: Eye prominent. Antennae similar length to each other. Antenna 2 peduncular articles robust but not elongated. Epistome and upper lip, in situ, directed straight down, perpendicular to the head. Lower lip outer plate entire. Mandible molar well developed and triturating; palp slender and three-articulate, article three distally rounded. Maxilla 1 palp moderately developed. Pereopods setose with plumose setae. Gnathopod 1 robust and sexually dimorphic, coxa slightly produced anteriorly, basis anteroventral lobe prominent and setose, propodus subrectangular, anterodistal setose lobe absent, palm acute and concave, defined by a very small rounded posterodistal tooth and a large robust seta; dactylus shorter than palm. Gnathopod 2 slender and slightly longer than gnathopod 1, sexually dimorphic; basis anteroventral lobe medium sized and setose; carpus subovoid; propodus subequal in length to carpus, propodus narrow, anterodistal lobe absent, palm acute, midpalmar tooth/corner present, defining posterodistal tooth absent, robust seta present; dactylus shorter than palm length. Pereopods 3 and 4 similar in size and shape, basis expanded and glandular; merus slightly expanded, lobe present. Pereopod 5 basis ovoid; distal articles slender; propodus weakly prehensile. Pereopods 6-7 similar lengths; merus and carpus broader than propodus; propodus weakly prehensile.

Epimeron 3 posteroventral corner rounded without tooth. Uropod 1, in situ, reaching to the end of uropod 2, peduncle distoventral spur absent. Uropod 2 peduncle rounded lateral distoventral process absent. Uropod 3 broad, peduncle with two distal robust setae; rami very short, outer ramus with two recurved robust setae, patch of denticles; inner ramus with just slender distal setae. Telson subrectangular, cusps absent, light denticles present, with lateral and apical setae.

Female. Similar to male except for gnathopod 1 merus lobe reduced and weakly setose, carpus more slender than male and less setose and subequal in length to the propodus; propodus narrow, weakly setose, palm convex, not sculptured.

**Etymology.** Named plumosa, a derivative of the latin plumosus meaning feathered, referring to the feathered nature of majority of the setae present on the type material.



Figure 1. Exampithoe plumosa sp. n., holotype, male, 10 mm, NIWA 121270, Otago Harbour, New Zealand.

**Remarks.** This is an interesting species for a couple of reasons. Firstly, it is the first record of this genus from the South Island of New Zealand. The only other Exampithoe species recorded from New Zealand is Exampithoe taylori Hughes & Peart, 2015. These two species are recorded from almost opposite ends of the country with over 1000 km between them and situated on water bodies influenced by different currents and geophysical history. Though the two species have some similarities, such as the shape of gnathopod 2 propodus (narrow, palm with a subquadrate midmedial tooth), the shape of the lower lip outer plate (entire), similar setation and shape of uropod 3 (broad peduncle, small rami, 2 distal peduncular robust setae, 1 marginal robust seta), there are also a number strong differences that give the necessity of these being separate species. These differences include: the shape and length of the antennae (A1 and A2 similar length in E. plumosa sp. n. A1 shorter than A2 in E. taylori. Antenna 2 peduncular articles are robust but not elongated in E. plumosa sp. n. and are robust but considerably elongated in E. taylori); the shape and size of gnathopod 1 (robust with shortened articles, propodus ovoid to subrectangular, palm excavate with small posterodistal tooth, subquadrate predactylus tooth and large defining robust seta in E. plumosa n.sp. and narrow with elongated articles, propodus subrectangular, palm convex, no defining tooth or predactylus tooth or robust seta in E. taylori). The other main difference between the two species is the majority of the setae on every appendage of *E. plumosa* are feathered (plumose) giving this animal a strongly fuzzy look. Whilst *E. taylori* has numerous setae on the appendages, very few are plumose.



Figure 2. *Exampithoe plumosa* sp. n. holotype, male, 10 mm, NIWA 121270, Otago Harbour, New Zealand. Scale bars: 0.5 mm (A1-2); 0.1 mm (MX1, MXP, LL); 0.2 mm (UL, MD).



**Figure 3.** *Exampithoe plumosa* sp. n. holotype, male, 10 mm, NIWA 121270, paratype, female, 7 mm, NIWA 121269, Otago Harbour, New Zealand. Scale bars: 0.5 mm (male **G1-2**); 0.2 mm (female **G1-2**).



Figure 4. *Exampithoe plumosa* sp. n. holotype, male, 10 mm, NIWA 121270, Otago Harbour, New Zealand. Scale bars: 0.5 mm (**P3-7**); 0.2 mm (**U1-3,T**).

The second interesting aspect of the discovery of this species is that the specimens were collected from a kelp raft. While many organisms can be found on floating or rafting macroalgae, ampithoids are only occasionally recorded rafting (Thiel and Gutow 2005) but are not obligate rafters. As there are no other records of this species, it cannot be inferred whether it is an obligate rafter or not. The kelp was determined to have been drifting in the water for around five weeks (determined by the stage of goose barnacle

settlement - Waters et al. in press) and probably originating from southern New Zealand. This is the first record of an ampithoid in this area of New Zealand.

**Distribution.** Only known from the type locality, Otago Harbour, South Island, New Zealand.

### Exampithoe taylori Hughes & Peart, 2015

*Exampithoe taylori* Hughes & Peart, 2015: 563–566, figs 4–6. *Ampithoe* sp. – J.L Barnard, 1972: 45, fig. 15.

**Type material.** Holotype: male, 11.5 mm, 4 slides, NIWA 94663, Leigh, New Zealand, 36°17'28.76"S, 174°48'10.81"E, coll. R. Taylor, 16 September 2002. Paratypes: 2 males, AM P. 88414, same location as holotype.

Additional material examined. NIWA 94664, 1 male, 9 mm; NIWA 94665, 1 female, 8 mm; NIWA 94666, 1 female, 7 mm; NIWA 94667, 1 female, 10 mm; NIWA 94668, 1 female, 8 mm; NIWA 94669, 1 male, 8 mm; NIWA 94670, 1 male, 7 mm; AM P.92556, 11 females, 15 males, Nordic Cove, Leigh, New Zealand, 36°17'28.76"S, 174°48'10.81"E, 1–3 m, on *Dictyota kunthii*, coll. Richard Taylor, 31 Dec 2009. NIWA 7022, 1 female, 6.4 mm, station E979, Huaroa Pt. Whangaparaoa Peninsula, Auckland Province, New Zealand, 36.59°S 175.84°E, shore collection, coll. J.L. Barnard, 16 Feb 1968.

Diagnosis. Male: Eye prominent. Antenna 1 slightly shorter than antenna 2. Epistome and upper lip, in situ, directed straight down, perpendicular to the head. Lower lip outer plate entire. Mandible molar reasonably well developed and triturating, palp slender and three-articulate, article three distally rounded. Maxilla 1 palp well developed. Pereopods weakly setose. Gnathopod 1 weakly sexually dimorphic; coxa slightly produced anteriorly; basis anteroventral lobe prominent; propodus subrectangular, anterodistal setose lobe absent, palm acute and convex, defining tooth and robust seta absent; dactylus subequal in length to palm. Gnathopod 2 slightly narrower and slightly longer than gnathopod 1, weakly sexually dimorphic; basis anteroventral lobe medium sized and setose; carpus elongated, subtriangular; propodus shorter than carpus, propodus narrow, anterodistal lobe absent, palm acute and convex, without defining tooth, but with a defining robust seta; dactylus subequal in length to palm. Pereopods 3 and 4 similar in size and shape; basis expanded and glandular; merus slightly expanded, lobe small and reduced. Pereopod 5 basis ovoid; distal articles slender; propodus weakly prehensile. Pereopods 6 and 7 similar in shape to each other, increasing in length; distal articles slender; propodus weakly prehensile. Epimeron 3 posteroventral corner rounded without tooth. Uropod 1, in situ, reaching to the end of uropod 2, peduncle distoventral spur absent. Uropod 2 peduncle without rounded lateral distoventral process. Uropod 3 broad, peduncle with two distal robust setae and 1 marginal robust seta; rami short; outer ramus with two recurved robust setae, lateral patch of denticles; inner ramus with robust and

slender distal setae. Telson subtriangular with reduced, small cusps, denticles absent, with lateral and apical setae.

Female: Similar to male.

**Remarks.** Known from northern New Zealand only. This species has similarities, but more significant differences to *E. plumosa* sp. n. These differences are discussed in the remarks above. Material examined from J.L. Barnard (1972) identified as *Ampithoe* sp. has been examined and identified as *Exampithoe taylori*.

Distribution. Leigh and Whangaparaoa, North Island, New Zealand.

### Pseudopleonexes Conlan, 1982

*Pseudopleonexes evensis* sp. n. http://zoobank.org/29497044-6CC3-4D94-BC16-6BB85E6C0404 Figs 5–7

Ampithoe (Pleonexes) lessoniae.—Barnard 1972: 44, figs 13–14.

**Type material.** Holotype, 9 mm, male, NIWA 121291, from algal washings, 0. 5 m, Eve Bay, Wellington, New Zealand, 41°19'58"S, 174°49'39"E, coll. R. Peart and J. Peart, 29 Nov 2016.

Paratype, female, 7 mm, NIWA 121292. Paratypes, female and male, ZMH K-46614, Same collection data as the holotype.

**Other material examined.** 6 specimens, male, female and juveniles, NIWA 121894, same collection data as the holotype. Male, 1 specimen, NIWA 7024, E797, from intertidal algal washings, 0.5 m, Huaroa Pt, Whangaparaoa, New Zealand, 36°35.7'S 175°50.14'E, coll J. L. Barnard, 16 Feb 1968.

Diagnosis. Male: eye prominent. Antennae damaged in type material (Barnard, 1972 material antenna 1 longer than antenna 2). Epistome and upper lip, in situ, directed posteriorly at an angle of around 45°. Lower lip outer plate weakly notched, lobes of even size. Mandibular molar reduced and triturating, palp reduced, 2 articles, article 2 distally rounded. Maxilla 1 palp 2-articulate, reduced and slender. Gnathopods densely setose, percopods weakly setose. Gnathopod 1 slender, sexually dimorphic; coxa slightly produced anteroventrally, basis anterodistal lobe medium and slightly upturned, bearing three slender setae; propodus subrectangular and narrow, with a strongly setose anterodistal lobe, palm acute, short, entire, without posterodistal tooth defining palm, without defining robust seta; dactylus greatly overreaching palm. Gnathopod 2 robust and longer than gnathopod 1, with long, dense simple setae on margins; basis anterodistal lobe medium and rounded, with four slender to robust setae on lobe margin, five robust setae on anterior margin of basis; carpus subtriangular; propodus longer than carpus; propodus broad, ovoid, produced into an anterodistally setose lobe; palm acute, excavate, with small subacute posterodistal tooth defining palm, with one defining robust seta; dactylus subequal in length to palm. Pereopods



Figure 5. Pseudopleonexes evensis sp. n. holotype, 9 mm, male, NIWA 121291, Eve Bay, Wellington, New Zealand.

3–4 similar in size and shape; basis expanded and glandular; merus expanded to form an acute lobe. Pereopod 5 basis circular, distal articles broad, propodus prehensile. Pereopod 6 shorter than pereopod 7, merus and carpus similar width to propodus, propodus prehensile. Epimeron 3 posteroventral corner rounded, no tooth. Uropod 1, in situ, reaching to end of uropod 2 peduncle; peduncle distoventral spur absent. Uropod 2 peduncle with rounded lateral distoventral process. Uropod 3 broad, peduncle with distal robust setae absent, rami short; outer ramus with two recurved robust setae; patch of denticles; inner ramus with just slender distal setae. Telson subtriangular, with two large recurved cusps and with 4 slender setae per lobe.

Female. Similar to the male except for gnathopod 1 basis anteroventral lobe reduced, merus is weakly setose; carpus shorter than propodus, carpal lobe slightly truncated; propodus weakly setose, anterodistal lobe reduced. Gnathopod 2 weakly setose, carpus shorter than propodus, carpal lobe rounded; propodus anterodistal lobe reduced, palm weakly excavate, posterodistal tooth reduced. Uropod 2 peduncle laterodistal projection absent.

**Remarks.** This relatively rare species can be aligned to J.L. Barnard's (1972) material and can be identified by the shape of the gnathopods 1 and 2 propodi and palms. The other interesting feature which when first observed in Barnard's description is the reduced mandibular palp. When the Eve Bay material was collected and dissected it was found to have a very similar mandibular form. If this is a valid character, along with the distinct presence of a setose anterodistal lobe on each of the gnathopod 1 and



**Figure 6.** *Pseudopleonexes evensis* sp. n. Holotype, 9 mm, male, NIWA 121291, Eve Bay, Wellington, New Zealand. Scale bars: 0.5 mm (mouthparts), 0.1 mm (**UI-3,T**).

2 propodi and the reduced, angled palm of gnathopod 1, validates this material as a new species.

Barnard (1972) described two males one 6.2 mm (E975), one 4.8 mm (E979), and mentioned in the description he only had two specimens and thought they maybe different species based on the presence/absence of the lobe on the uropod 2 peduncle (present on 6.2 mm male/absent on 4.8 mm male), the broadened articles of pereopod 5 (slightly broader than *P. lessoniae*) and the excavation of gnathopod 2 palm (more strongly excavated than *P. lessoniae*). He then mentions three stations where it was collected. The only material that has been able to be found is the 4.8 mm male and this


Figure 7. *Pseudopleonexes evensis* sp. n., holotype, 9 mm, male, NIWA 121291, Eve Bay, Wellington, New Zealand. Scale bars: 0.5 mm.

material (when examined) matches to *P. evensis* sp. n. and differs from *P. lessoniae* by the shape and structure of gnathopod 1 (consistent across sizes -9 mm length described here and the 4.8 mm male he described). The description notes that the uropod 2

peduncular process is absent, however when examined this character is obvioiusly there. The 6.2 mm material was not able to be located and so cannot be verified.

**Etymology.** The specific name is taken from the name of the type locality, Eve Bay. **Distribution.** North Island, New Zealand.

#### Pseudopleonexes lessoniae (Hurley, 1954)

Figs 9-12

Pleonexes lessoniae Hurley, 1954: 620-626, figs. 1-2.

*Pseudopleonexes lessoniae.*—Conlan 1982: 2020.—Just 2002: 31–40.—Peart 2006: 1–22.

not Ampithoe (Pleonexes) lessoniae.-Barnard 1972: 44, figs 13-14.

**Material examined.** Holotype: male, 9 mm, NIWA 121308, slide 90 Hurley collection, Island Bay, Wellington, New Zealand, 41°20'39.8"S, 174°46'25.9"E, on *Lessonia variegata*, coll. J.G. Gibbs, 1 August 1950.

Paratype: female, 5.75 mm, NIWA 12309, slide 91, Hurley collection, Island Bay, Wellington, New Zealand, 41°20'39.8"S, 174°46'25.9"E, on *Lessonia variegata*, coll. J.G. Gibbs, 1 August 1950.

All material (other than type material) collected by hand by M. Thiel & A.N. Lörz from *Macrocystis* sp. kelp, Breaker Bay, Wellington, 41.33° S, 174.83° E, 0–1 m between 30<sup>th</sup> Jan–1<sup>st</sup> Feb 2013: NIWA 96675–96677, 17 specimens; NIWA 96679, 5 specimens, NIWA 96683–96686, 9 specimens; NIWA 96688–96698, 27 specimens; NIWA 96700–96701, 3 specimens; NIWA 96792–96795, 10 specimens; NIWA 96797–96804, 42 specimens; NIWA 96810–96818, 59 specimens; NIWA 96820, 15 specimens; NIWA 96822, 14 specimens; NIWA 96824, 7 specimens; NIWA 96826, 1 specimen. NIWA 120146 on SEM stud

**Diagnosis.** Male: Eye absent (holotype), eyes prominent (additional material examined). Epistome and upper lip, in situ, directed posteriorly around 45°. Antenna 1 longer than antenna 2. Lower lip outer plate slightly notched, almost entire, margin sinusoidal to sometimes flat with larger, subacute corners. Mandible molar reduced and triturating, palp three-articulate, article three distally beaked. Maxilla 1 palp poorly developed and slender tipped with slender plumose setae. Gnathopods strongly setose. Pereopods weakly setose. Gnathopod 1 not sexually dimorphic; coxa not anteroventrally produced; basis anterodistal lobe reduced and rounded bearing on slender seta; propodus subrectangular, anterodistal setal lobe absent, palm transverse, entire, without midmedial tooth, with posterodistal tooth defining palm and one small defining robust seta; dactylus overreaching palm. Gnathopod 2 more robust and larger than gnathopod 1, sexually dimorphic, with long plumose setae on margins; basis anterodistal lobe large and rounded, bearing around 10 robust setae on the margin; carpus subtriangular; propodus longer than carpus; propodus broad, anterodistally setose lobe absent; palm acute, entire (sometimes slightly excavate), midpalmar tooth absent, with small suba-



Figure 8. *Pseudopleonexes evensis* sp. n., paratype, female, 7 mm, NIWA 121292, Eve Bay, Wellington, New Zealand. Scale bars: 0.5 mm (G1-2), 0.2 mm (U2).

cute posterodistal tooth defining palm, and one defining robust seta; dactylus subequal to palm. Pereopods 3–4 similar in size and shape, basis expanded and glandular; merus expanded and glandular, forming an acute lobe. Pereopod 5 basis circular, distal articles broad to slender (depending on size), propodus prehensile. Pereopod 6–7 increasing in length, merus and carpus similar widths to propodus (slightly wider), propodi prehensile. Epimeron 3 posteroventral corner rounded without tooth. Uropod 1, in situ, reaching only to the end of uropod 2 peduncle; peduncle distoventral spur absent. Uropod 2 peduncle with large rounded distolateral process. Uropod 3 broad, peduncle without distal robust setae, rami short, outer ramus with two strongly recurved robust setae, patch of denticles; inner ramus with just slender distal setae. Telson subtriangular with strongly recurved cusps, denticles absent, with one slender seta per lobe.



Figure 9. Pseudopleonexes lessoniae (Hurley, 1954), male, 9 mm, NIWA 96679, female, 8 mm, NIWA 96679, Breaker Bay Wellington. Scale bars: 0.5 mm.

**Remarks.** Described by Hurley (1954) from Wellington, New Zealand, this species is a small, robust amphipod dwelling in *Lessonia variegata*. Hurley (1954) described it as having similarities to members of the *Ampithoe* group, *Pleonexes*. When the



Figure 10. *Pseudopleonexes lessoniae* (Hurley, 1954), male, 9 mm, NIWA 96679, female, 8 mm, NIWA 96679, Breaker Bay Wellington. Scale bars: 0.5 mm.

genus *Pseudopleonexes* was constructed (Conlan, 1982), this species was placed as the type of the genus. It has strongly plumose, long setae on the gnathopod 2, a character represented in most of the other species in the genus. Hurley's types have been located, and are in the NIWA Invertebrate Collection (NIC).



**Figure 11.** *Pseudopleonexes lessoniae* (Hurley, 1954), male, 9 mm, NIWA 96679, female, 8 mm, NIWA 96679, Breaker Bay Wellington. Scale bars: 0.5 mm.



**Figure 12.** *Pseudopleonexes lessoniae* (Hurley, 1954), male, NIWA 120146, Breaker Bay, Wellington, New Zealand. **A** whole animal, habitus **B** close up of propodus and dactylus pereopod 6 **C** close up of pereon and coxa showing position pits on the surface **D** close up of Uropod 3 rami **E**, **F** magnification of the heart shaped (**E**) and butterfly shaped (**F**) pits on the coxae and pereon.

Barnard (1972) assigned two specimens from New Zealand as *P. lessoniae* however, examination of one of the specimens (the other is not locatable) and some confusion in the description indicate that these are not of this species. Comparison with freshly collected material indicates these should be treated as a new species (described above as *P. evensis* sp. n.). *Pseudopleonexes lessoniae* sensu stricto differs from *P. evensis* sp. n. by the absence of an anterodistal setose lobe on the propodi of gnathopods 1 and 2 (strongly present in *P. evensis* sp. n.) and the strongly transverse gnathopod 1 palm, also half the width of the propodus (acute, greatly reduced palm in *P. evensis* sp. n.).

The material described here is from Breaker Bay, very close to the type locality of Island Bay (4.5 km ENE) and was very abundant in *Macrocystis* sp. and until molecular examination is carried out is placed with *P. lessoniae* (Hurley, 1954). The main differences involve the apparent presence/absence of the eye and presence of butterfly and heart-shaped pits and fine hairs covering the pereon and pereopods of the recently collected material (fig. 12).

Distribution. Wellington area, New Zealand.

#### Sunamphitoe Spence Bate, 1857

#### Sunamphitoe aorangi (J.L. Barnard, 1972)

Ampithoe aorangi J.L. Barnard, 1972: 27, 37, figs 8, 9 (part, not 10a–e) Peramphithoe aorangi.—Shin et al. 2015: 261–264. Sunamphitoe aorangi.—Peart, 2017: 308 Not Peramphithoe aorangi.—Hughes and Peart 2014: 93–95, fig. 61.

**Material examined.** Holotype, male, 5.3 mm, NIWA 798, intertidal wash of algae and their rhizomes, Eve Bay, off Strathmore Park, Wellington, New Zealand, 41°19.8'S 174°49.8'E, NZOI Sta E966, coll. J.L. Barnard, 5 Nov 1968.

Diagnosis. Male: Eye prominent. Antenna 1 longer than antenna 2. Epistome and upper lip, in situ, directed straight down, perpendicular to the head. Lower lip outer plate notched, lobes of equal size. Mandible molar triturating, palp with 3 articles, article three distal margin rounded. Maxilla 1 palp well developed. Gnathopods and percopods weakly setose. Gnathopod 1 coxa slightly produced anteroventrally to form a slight rounded tooth; basis anteroventral lobe reduced and rounded bearing one slender seta; propodus subrectangular, anterodistal setose lobe absent; palm transverse defined by a small robust seta, posterodistal tooth absent; dactylus overreaching the palm. Gnathopod 2 more robust and slightly larger than gnathopod 1, sexually dimorphic, basis anteroventral lobe reduced and rounded with very small setae; carpus subtriangular; propodus longer than carpus; propodus broad, ovoid, anterodistal lobe absent, palm slightly acute (close to transverse), entire, defining posterodistal tooth absent, defining robust seta present; dactylus overreaching the palm. Pereopods 3-4 similar in size and shape; basis expanded and glandular; merus expanded and glandular, lobe subacute. Pereopod 5 basis ovoid to circular, distal articles slightly broadened, propodus weakly prehensile. Pereopods 6–7 similar size, distal articles slender, propodus weakly prehensile. Epimeron 3 posteroventral corner broadly rounded, tooth absent. Uropod 1, in situ, reaching to the end of uropod 2 rami, peduncle acute distoventral spur present. Uropod 2 peduncle rounded lateral distoventral process absent. Uropod 3 narrow, peduncle with distal robust setae absent; outer ramus two strongly recurved robust setae,

with patch of lateral denticles; inner ramus with one distal robust seta and slender setae. Telson subtriangular, apical cusps reduced and rounded, no denticles, with both lateral and apical slender setae.

Female. Not documented.

**Remarks.** This species has recently been redescribed (Peart 2017) and the original material confusion resolved. The differences between this species and the recently described only other species from New Zealand (*S. mixtura* Peart, 2017) are detailed in Table 1 of that publication.

Distribution. Wellington, New Zealand.

#### Sunamphitoe mixtura Peart, 2017

Sunamphitoe mixtura Peart, 2017: 326 Ampithoe aorangi.—Barnard 1972: 37 (part, sta. E978), fig. 10a–e.

**Type material.** Holotype: male, 7.8 mm, NIWA 892, small high rock pools, in surf splash zone, lined with filamentous brown alga, Huaroa Point, Whangaparaoa Peninsula (Auckland Province), NZOI Sta. stn E978, coll. J.L. Barnard, 16 Feb 1968.

Diagnosis. Male. Epistome and upper lip, in situ, directed straight down, perpendicular to the head. Lower lip outer plate notched, lobes of equal height. Mandibular molar well developed, triturating; palp with 3 articles, article three rounded distally. Maxilla 1 palp well developed. Gnathopods and pereopods weakly setose. Gnathopod 1 coxa not produced anteroventrally, basis anteroventral lobe medium in size and rounded, bearing three small setae; propodus subrectangular, not produced to form an anterodistal lobe; palm transverse, not defined by a posterodistal tooth, but with a small posterodistal robust seta; dactylus overreaching palm. Gnathopod 2 sexually dimorphic, larger than and more expanded than gnathopod 1; basis anterodistal lobe reduced and rounded bearing three slender setae; carpus very short and subtriangular; propodus much longer than carpus; propodus subtriangular (expanded proximally, narrow distally), anterodistal lobe absent; palm acute, excavate, sculptured, defining posterodistal tooth and robust seta present; dactylus subequal in length to palm. Pereopods 3 basis expanded and glandular; merus expanded with subacute lobe. Perepods 4-7 missing. Epimeron 3 not documented. Uropod 1, in situ, reaching to the end of uropod 2; peduncle with large acute distoventral spur. Uropod 2 peduncle rounded lateral distoventral process absent. Uropod 3 narrow, rami small, peduncle with one distal robust seta; outer ramus with two large recurved robust setae, with patch of lateral denticles; inner ramus with three robust setae and many slender setae. Telson subtriangular, apical cusps small, reduced and rounded, with apical and lateral setae and lateral denticles.

## Key to the New Zealand species of the family Ampithoidae

1	Uropod 1 peduncle acute distoventral spur present
_	Uropod 1 peduncle distoventral spur absent
2	Gnathopod 2 propodus subtriangular, greatly expanded proximally, narrow
	distally, palm excavate Sunamphitoe mixtura Peart, 2017
_	Gnathopod 2 propodus subovoid, broad evenly along length, palm entire
3	Gnathopod 1 palm greatly acute
_	Gnathopod 1 palm slightly acute or transverse
4	Gnathopod 2 propodus subrectangular and narrow
_	Gnathopod 2 subovoid and broad Ampithoe hinatore J.L. Barnard, 1972
5	Gnathopod 1 propodus broad proximally and narrow distally, palm excavate,
	prepalmar tooth present, large defining robust seta present
	<i>Exampithoe plumosa</i> sp. n.
_	Gnathopod 1 propodus elongated and narrow, palm entire, no defining ro-
	bust seta Exampithoe taylori Hughes & Peart, 2015
6	Gnathopods 1 and 2 propodi with setose anterodistal lobes
	Pseudopleonexes evensis sp. n.
_	Gnathopods 1 and 2 propodi without setose anterodistal lobes

## Acknowledgements

The authors would like to thank Martin Thiel for instigating and undertaking the project and field work along with ANL. Thanks also to Jonathan Waters for making the *Exampithoe* material available. Thank you to Sadie Mills, Kareen Schnabel and Caroline Chin for curating the material and making the NIWA Invertebrate Collection material available. Thank you to Erika Mackay for speedily and beautifully constructing the SEM plate. Renate Walter (Zoological Museum Hamburg) kindly assisted with the scanning electron microscope.

## References

- Audouin V (1826) Explication sommaire des planches de crustaces de l'Egypte et de la Syrie, publiées par Jules-Cesar Savigny, membre de l'Institut; offrant un exposé des caractères naturels des genres, avec la distinction des espèces. Description de l'Egypte, Histoire Naturelle 1: 77–98.
- Barnard JL (1970) Sublittoral gammaridea (Amphipoda) of the Hawaiian Islands. Smithsonian contributions to Zoology 34: 1–286. https://doi.org/10.5479/si.00810282.34

- Barnard JL (1972) The marine fauna of New Zealand: algae-living littoral Gammaridea (Crustacea Amphipoda). Memoir of the New Zealand Oceanographic Institute 62: 7–216.
- Barnard KH (1925) Contributions to the crustacean fauna of South Africa. No. 8. Further additions to the list of Amphipoda. Annals of the South African Museum 20: 319–380.
- Boeck A (1870) Crustacea amphipoda borealia et arctica. Forhandlinger i Videnskabs-Selskabet i Christiania. 1870: 83–280.
- Conlan KE (1982) Revision of the gammaridean amphipod family Ampithoidae using numerical analytical methods. Canadian Journal of Zoology 60: 2015–2027. https://doi.org/10.1139/z82-259
- De Broyer C, Lowry JK, Jazdzewski K, Robert H (2007) Catalogue of the Gammaridean and Corophiidean Amphipoda (Crustacea) of the Southern Ocean, with distribution and ecological data. In: De Broyer C (Ed) Census of Antarctic Marine Life: Synopsis of the Amphipoda of the Southern Ocean. Vol. I. Bulletin de l'Institut royal des Sciences naturelles de Belgique, Biologie 77, suppl. 1: 1–325.
- Fenwick GD (1976) The effect of wave exposure on the amphipod fauna of the alga *Caulerpa brownia*. Journal of Experimental Marine Biology and Ecology 25(1): 1–18. https://doi.org/10.1016/0022-0981(76)90072-1
- Horton T, De Broyer C, Costello M, Bellan-Santini D (2017) Ampithoidae Boeck, 1871. In: Horton T, Lowry J, De Broyer C, Bellan-Santini D, Coleman CO, Daneliya M, Dauvin J-C, Fišer C, Gasca R, Grabowski M, Guerra-García JM, Hendrycks E, Holsinger J, Hughes L, Jaume D, Jazdzewski K, Just J, Kamaltynov RM, Kim Y-H, King R, Krapp-Schickel T, LeCroy S, Lörz A-N, Senna AR, Serejo C, Sket B, Tandberg AH, Thomas J, Thurston M, Vader W, Väinölä R, Vonk R, White K, Zeidler W (Eds) World Amphipoda Database. Accessed through: World Register of Marine Species at http://www.marinespecies.org/aphia. php?p=taxdetails&id=101366 [accessed on 2017-05-17]
- Hughes LE, Peart RA (2014) New species and new records of Ampithoidae (Peracarida: Amphipoda) from Australian waters. Zootaxa 3719 (1): 1–102. https://doi.org/10.11646/ zootaxa.3719.1.1
- Hughes LE, Peart RA (2015) Three new species of *Exampithoe* from Australia and New Zealand (Ampithoidae: Amphipoda: Crustacea). Zootaxa 3918 (4): 559–570. https://doi.org/10.11646/zootaxa.3918.4.5
- Hurley DE (1954) Studies on the New Zealand Amphipodan Fauna No. 5 *Pleonexes lessoniae*, a new species of the family Ampithoidae. Transactions of the Royal Society of New Zealand, 81(4): 619–626.
- Just J (2002) Review of *Pseudopleonexes* Conlan, 1982, with a new species from Australia (Crustacea: Amphipoda: Ampithoidae). Records of the Australian Museum 54(1): 31–40. https://doi.org/10.3853/j.0067-1975.54.2002.1361
- Lowry JK (1974) Key and Checklist to the Gammaridean Amphipods of Kaikoura. Mauri Ora 2: 95–130.
- Peart RA (2006) A revision of *Pseudopleonexes* Conlan, 1982 (Crustacea: Amphipoda: Ampithoidae) with description of three new species from Australia. Zootaxa 1344: 1–22.

- Peart RA (2017) Analysis of the genus *Sunamphitoe* Spence Bate, 1857 (Amphipoda: Ampithoidae) with descriptions of eight new species. Zootaxa 4269(3): 301–345. https://doi. org/10.11646/zootaxa.4269.3.1
- Shin M-H, Coleman CO, Hong JS, Kim W (2015) A new species of *Peramphithoe* (Amphipoda: Ampithoidae) from South Korea, with morphological diagnoses of the world cogeneric species. Journal of Crustacean Biology 35: 255–270. https://doi.org/10.1163/1937240X-00002323
- Spence Bate C (1857) A synopsis of the British edriophthalmous Crustacea. Annals and Magazine of Natural History (Series 2) 19: 135–152.
- Thiel M, Gutow L (2005) The ecology of rafting in the marine environment. I The floating substrata. In: Gibson RN, Atkinson RJA, Gordon JDM (Eds) Oceanography and Marine Biology: An annual review. CRC Press 42, 181–263.
- Waters JM, Nikula R, Ahyong ST, Arango CP, Bruce NL, Marshall BA, Willan RC, Spencer HG, Fraser CI (in press) Rafting of a diverse coastal benthic assemblage in southern New Zealand. New Zealand Journal of Marine and Freshwater Research.

RESEARCH ARTICLE



# Two new species of the genus *Dahlica* Enderlein (Lepidoptera, Psychidae) from Korea

Seung Jin Roh<sup>1</sup>, Bong-Woo Lee<sup>1</sup>, Bong-Kyu Byun<sup>2</sup>

l Division of Forest Biodiversity, Korea National Arboretum, Pocheon, South Korea **2** Department of Biological Science and Biotechnology, Hannam University, Daejeon, South Korea

Corresponding author: Bong-Kyu Byun (bkbyun@hnu.kr)

Academic editor: <i>E. van Nieukerken</i>   Received 4 September 2017   Accepted 7 December 2017   Published 26 January 2018
http://zoobank.org/57A16921-EE7A-4881-8C62-81DDFF247B23

**Citation:** Roh SJ, Lee B-W, Byun B-K (2018) Two new species of the genus *Dahlica* Enderlein (Lepidoptera, Psychidae) from Korea. ZooKeys 733: 49–64. https://doi.org/10.3897/zookeys.733.20793

#### Abstract

The genus *Dahlica* Enderlein, 1912 is reported for the first time from Korea with two new species: *Dahlica* (*Dahlica*) somae Roh & Byun, **sp. n.** and *Dahlica* (*Dahlica*) ochrostigma Roh & Byun, **sp. n.** Adults and genitalia are illustrated, and DNA barcodes for precise identification of the species are also provided.

#### Keywords

bagworms, DNA barcode, Naryciinae, new species, Psychidae

## Introduction

The family Psychidae is a medium-sized family of moths consisting of 241 named genera and 1,350 species (Sobczyk 2011; van Nieukerken et al. 2011). Phylogenetically, Psychidae belong to the oldest clades of the suborder Ditrysia in the order Lepidoptera, and have usually been placed in the superfamily Tineoidea (Davis and Robinson 1998),

with Eriocottidae, Tineidae, Meessiidae, and Dryadaulidae as phylogenetically allied groups (Mutanen et al. 2010; Regier et al. 2015). Most species of Psychidae produce characteristic cases or bags at different larval stages (Sugimoto 2009a, 2009b), which gives rise to their common name, bagworms. Parthenogenesis is known in several species of the genus *Dahlica* Enderlein, 1912 in the Naryciinae (Grapputo et al. 2005; Elzinga et al. 2013). Identification of these species and classification of the females based on morphological and ecological characters alone is difficult (Grapputo et al. 2005; Elzinga et al. 2013). In Korea, Roh et al. (2016) reviewed the nine known species including a new species, *Psyche yeongwolensis* Byun & Roh, 2016 and recorded a species new for the country, *Proutia maculatella* Saigusa & Sugimoto, 2014. Later, Roh and Byun (2016) recorded *Ceratosticha leptodeta* Meyrick, 1935 new for Korea. Recently, three more species were reported: *Bacotia sakabei* Seino, 1981, (Roh and Byun 2017a), *Bruandella niphonica* (Hori, 1926), and *Proutia nigra* Saigusa & Sugimoto, 2014 (Roh and Byun 2017b). Consequently, 13 species in total are now known from Korea.

The genus *Dahlica* was based on the type species *Dahlica larviformis* Enderlein, 1912 by Enderlein in 1912 (Sobczyk 2011). The members of *Dahlica* are superficially similar to *Siederia* Meier, 1957 (Grapputo et al. 2005), but can be distinguished from the latter by the absence of an epiphysis on the fore-tibia of the male (Herrmann 1988; Herrmann and Weidlich 1999; Rekelj et al. 2014; Arnscheid 2016), the absence of the medial cell in the fore- and hindwings, presence of accessory cells, and six veins arising from the discoidal cell of the hindwing (Rekelj and Predovnik 2014).

Meier (1958) and Sieder (1953) proposed to divide *Dahlica* in various subgenera, which were later raised to genus. Recently Arnscheid and Weidlich (2017) reviewed the five allied genera, *Dahlica, Siederia, Brevantennia* Sieder, 1953, *Postsolenobia* Meyer, 1958, and *Praesolenobia* Sieder, 1955, and decided on the basis of the venation of the male hindwings, male forewing scale morphology, presence of an epiphysis in the males, the structure of reproductive organs, and the female antennae to sank these genera again as subgenera of *Dahlica*. They diagnosed the subgenus *Dahlica* by the following characters: the absence of an epiphysis, presence of six veins from the hindwing discal cell, and the long female antennae, with more than eleven segments (Arnscheid and Weidlich 2017).

Females of the genus *Dahlica* are unable to fly because of their degenerate wings (Sauter and Hättenschwiler 1999). The larvae feed on moss, algae, and lichens, which are attached to walls or the bark of trees via a sac constructed of small sand particles (Sauter and Hättenschwiler 1999; Sugimoto 2009a; Arnscheid and Weidlich 2017).

In total, 42 species of the subgenus *Dahlica* have been reported worldwide and are distributed throughout the Palaearctic region in Europe (41 species) and Asia (one species) (Sobczyk 2011; Arnscheid and Weidlich 2017).

In this study, *Dahlica* (*Dahlica*) somae sp. n. and *D*. (*D*.) ochrostigma sp. n. are described as new species and the genus *Dahlica* is reported for the first time from Korea. All available information is presented, including the collection locations, microhabitats, and illustrations of adults and their genitalia. DNA barcodes are also provided for precise identification of each species.

#### Materials and methods

The material examined in this study is preserved in the Systematic Entomology Laboratory, Hannam University (**SEL/HNU**), Daejeon, Korea, and the Entomological Collection of the Korea National Arboretum, Pocheon, Korea (**KNAE**). Specimens were dissected and examined after mounting on slide glass; male genitalia and wing scales in 80 % glycerol solution, females in euparal solution and wing venation on dried condition. Photographs of adults and genitalia were taken using a PAXcam digital camera (PAXcam Microscope Cameras Co., Chicago, IL, USA) attached to a Carl Zeiss Axio Imager A1 microscope (Carl Zeiss Ltd., Cambridge, MA, USA).

Terminology and morphological characters of the adult, wing venation, and genitalia follows Dierl (1964), Kristensen (2003), and Arnscheid and Weidlich (2017) (Figs 1–4) and the terminology for forewing scales (class 1 to 6) follows Sauter (1956). The set-up of the data matrix for morphological characters of the genus *Dahlica* follows Arnscheid (2016) (Table 2).

Genomic DNA was extracted from the legs of dried specimen for males and thorax parts of immersion specimen for females, preserved in 100% alcohol using a Genomic Cell/Tissue Spin Mini Kit (Mbiotech, Inc., Hanam, Korea), according to the manufacturer's protocol. A total of six specimens were sequenced for, the 658 bp fragment of the mitochondrial cytochrome c oxidase I (COI) gene, the DNA barcode, was amplified using the primer pair LepF1 and LepR1 (Hebert et al. 2004). PCR conditions for amplification followed the manufacturer's protocol (Platinum Taq, Invitrogen, Carlsbad City, CA, USA). Amplicons were purified using the QIAquick<sup>®</sup> PCR purification kit (QIAGEN, Inc.) and directly sequenced at Genotech Corp. (Yuseong-gu, Daejeon, Korea). Contigs were assembled using CodonCode aligner version 2.0.6 (CodonCode Co., Centerville City, MA, USA) and were aligned using MAFFT (Katoh and Toh 2008).

The new barcodes were compared to 18 DNA barcodes of the genera *Dahlica* and *Narycia* downloaded from GenBank (National Center for Biotechnology Information, USA, http://www.ncbi.nlm.nih.gov/) (Table 1). A neighbor-joining (NJ) analysis was performed with MEGA 6.0 (Tamura et al 2013) under the K2P model for nucleotide substitutions. Successful sequences were uploaded to BOLD systems (project. KNAE) and submitted to GenBank (Table 1).

#### Systematic accounts

#### Dahlica Enderlein, 1912

Subgenus *Dahlica* Enderlein, 1912 *Dahlica Enderlein* 1912: 264.

Type species. Dahlica larviformis Enderlein, 1912: 264 by monotypy.



**Figures 1–4.** Terminology of morphological characters. **I** Male (Dierl (1964), Arnscheid and Weidlich (2017)) **2** Male genitalia, dorso-ventral part (Dierl (1964), Kristensen (2003) and Arnscheid and Weidlich (2017)) **3** Ditto, lateral part (Dierl (1964), Kristensen (2003) and Arnscheid and Weidlich (2017)) **4** Female (Arnscheid and Weidlich (2017)).

## Key to the males of Dahlica in Korea

1	Hindwing M2 and M3, originate at apical	corner of posterior part of discoi-
	dal cell (Fig. 28), dorsum of genitalia gently	y curved to apical part and harpe
	hooked (Fig. 11)	
_	Hindwing $M_3$ stalked at 1/4 $M_2$ , dorsum st	rongly arched to apical part (Fig.
	29) and harpe needle shape (Fig. 18)	D. (D.) ochrostigma sp. n.

Scientific name	Country	BIN number	Accession number (GenBank)	Basepair length
Dahlica (Dahlica) somae sp. n.	Korea	BOLD:ADJ8202	MF508656	658
D. (D.) somae	Korea	BOLD:ADJ8201	MF664099	658
D. (D.) somae	Korea	BOLD:ADJ8201	MF664100	658
D. (D.) ochrostigma sp. n.	Korea	BOLD:ADK4708	MF508657	658
D. (D.) ochrostigma	Korea	BOLD:ADK8063	MF664101	658
D. (D.) ochrostigma	Korea	BOLD:ADK8063	MF664102	658
D. (D.) triquetrella (Hübner)	Canada	_	KR941436	591
D. (D.) triquetrella	Switzerland	-	KX045622	658
D. (D.) triquetrella	Slovenia	-	KX045823	658
D. (D.) lichenella (Linnaeus)	Canada	-	KR941275	591
D. (D.) fennicella (Suomolainen)	Finland	-	JX307942	657
D. (D.) lazuri (Clerck)	Finland	_	JX307894	657
D. (D.) goltella Rekelj & Predovnik	Slovenia	_	KX045455	658
D. (D.) charlottae (Meier)	Finland	_	JX307874	657
D. (D.) parthenogenesis (Saigusa)	Japan	-	LC094189	665
Dahlica (Postsolenobia) juliella (Rebel)	Slovenia	_	KX047137	658
Dahlica (Siederia) listerella (Linnaeus)	Japan	-	LC094179	665
D. (S.) listerella	Austria	-	KP150244	658
D. (S.) listerella	Finland	_	KJ192386	658
D. (S.) rupicolella (Sauter)	Finland	_	KJ192382	658
Dahlica (Bevantennia) adriatica (Rebel)	Slovenia	_	KX045214	658
Narycia emikoae Niitsu, Jinbo & Nasu	Japan	_	LC160295	658
Narycia duplicella (Goeze)	Slovenia	_	KX045830	658
N. duplicella	Belgium	_	KC305219	658

Table 1. Species with DNA barcodes and GenBank accession numbers used in this study.

#### Dahlica (Dahlica) somae Roh & Byun, sp. n.

http://zoobank.org/E35CEE22-4005-4581-AFAD-DEB937241716 Figs 5–12, 24–28, 32, 33

**Type material.** *Holotype.* ♂, **Korea**: Daejeon, Mt. Heungnyongsan, 15.ii.2015, S.J. Roh & D.S. Kim, genitalia mounted on 80% glycerol solution, genitalia No. KNAESJ01, scales of forewing mounted on 80% glycerol solution, scales of forewing No. KNAESSJ01, venation of forewing No. KNAEVSJ01, DNA barcode accession No. MF508656. Deposited at SEL/HNU.

*Paratypes.* 2♂, 1♀. Korea: 1♂ Daejeon, Isa-dong, 2.ii.2015, S.J. Roh, genitalia mounted on 80% glycerol solution, genitalia No. KNAESJ02, scales of forewing mounted on 80% glycerol solution, scales of forewing No. KNAESSJ02, venation of forewing No. KNAEVSJ02, DNA barcode accession No. MF664099; 1♂ Daejeon, Mt. Heungnyongsan, 6.iii.2017, S.J. Roh & D.S. Kim, genitalia mounted on 80% glycerol solution, genitalia No. KNAESJ03; 1♀ Mt. Heungnyongsan, 6.iii.2017, S.J. Roh & D.S. Kim, DNA barcode accession No. MF664100. Deposited at SEL/HNU.

Species	Male wingspan	Scales (classes)	Hindwing venation (M <sub>2</sub> /M <sub>3</sub> )	Genitalia index
D. (D.) somae	12.3–13.4 mm.	2-4	free	1.46–1.56
D. (D.) ochrostigma	9.8–11.2 mm.	1-2	short stalked	0.79-1.08

Table 2. Data matrix for morphological characters (Arnscheid 2016) of Dahlica species in Korea.

**Diagnosis.** Male of this species is superficially similar to *D. triquetrella* (Hübner, 1813), but can be distinguished by a slightly longer transtilla and a relatively short ampulla of the male genitalia (lateral aspect). This species can be readily differentiated by the veins of the male hindwing; M2 and M3 originate at the apical corner of the posterior part of the discoidal cell. Female apophyses posteriores 1.75 times longer than apophyses anteriores.

Description. Adult. Male (Figs 5–12). Wingspan 12.3–13.4 mm (Table 2). Coloration and vestiture: Vertex of head roughly covered with grayish brown hairs. Thoracic notum covered with blackish brown hairs. Upper side of forewing: ground color grayish black; white spots present regularly; scales (Fig. 10) slightly narrow and evenly widened apically; apical margin usually produced into two to four laciniations (classes 2-4) (Table 2). Hindwing covered with grayish white scales; postmarginal part present with slight long shiny white hairs. Structure: head and compound eyes slightly large; ocelli absent. Antennae (Fig. 8) filiform, longer than 2/3 forewing. Forewing: slightly long and narrow; costa straight; termen shortly arched to posterior margin, discoidal cell 0.64 times as long as forewing; venation (Fig. 28) with nine veins, originating at the discoidal cell; accessory cell present; intercalary cell absent; Sc arising with 3/5 costa; R, and R<sub>3</sub> originating at corner of accessory cell; R<sub>4</sub> and R<sub>5</sub> fused and originating at apical corner of anterior part of the discoidal cell reaching to the apex; M<sub>1</sub> and M<sub>2</sub> parallel; M<sub>2</sub> and M<sub>3</sub> stalked at apical corner of posterior part of the discoidal cell; Cu, and Cu, parallel. Hindwing (Fig. 28): costa straight; discoidal cell 0.51 times as long as hindwing; Sc straight to 4/5 costa; R terminating at apex; M, and M, parallel, M<sub>2</sub> and M<sub>3</sub> originating at apical corner of posterior part of the discoidal cell (Table 2); Cu, and Cu, parallel to tornus. Legs: epiphysis absent (Fig. 9); femora and tibiae covered with brown hairs; tarsi covered with grayish brown scales.

*Female* (Figs 24–27). Adult 4.2 mm in length. Coloration: Head, meso-, and metanotum dark brown. Membranous areas of abdomen yellowish brown. Abdomen covered with light brown scales; corethrogyne densely covered with white hairs on ventral part only. Structure: apterous. Head and compound eyes small, antennae slightly developed with basal flagellomeres 17 segmented, bipectinated. Legs well developed with tarsi divided into four tarsomeres; hind legs present with apical spurs.

*Male genitalia* (Figs 11, 12). In lateral aspect. Genitalia index, 1.46–1.56 (Table 2). Dorsum gently curved. Saccus very short; ampulla narrow and short with club shape, setae present sparsely; harpe short with hooked shape; phallus slender and very long with whip shape. In dorso-ventral aspect, uncus slightly concave; gnathos and juxta absent; valva slightly narrow, apical part of valva densely covered.



Figures 5–12. Male of *Dahlica (Dahlica) somae*, holotype. 5 Male 6 Close-up of rightwing- pattern
7 Head, frontal view 8 Antenna 9 Absence of foreleg-tibia 10 Scales of forewing (slide No. KNAESSJ01)
11 Genitalia (slide No. KNAESJ01), lateral view 12 Ditto, dorso-ventral view.

*Female genitalia* (Fig. 27). Oviscapt and ostium bursae well sclerotized. Apophyses posteriores 1.75 times longer than apophyses anteriores, very slender. Sclerotizations of the seventh sternite present with bundle of hairs



Figures 13–19. Male of *Dahlica (Dahlica) ochrostigma*. 13 Male, holotype 14 Close-up of rightwingpattern, Paratype (Korea, Gangwon-do, Taebaek-si, Changjuk-dong, GW, 6.iii.2015) 15 Antenna, ditto 16 Scales of forewing, holotype (slide No. KNAESSJ03) 17 Absence of foreleg-tibia, holotype 18 Genitalia, holotype (slide No. KNAESJ04), lateral view 19 Ditto, dorso-ventral view.

*Larval case* (Figs 32, 33). Length 4.0 mm. Larvae build their cases by putting together small sand particles, forming oval-shaped cases rather than angular cases. **Distribution.** Korea.



Figures 20–27. Females. 20 *Dahlica (Dahlica) ochrostigma* preserved in 70% ethanol, paratype (Korea, Gangwon-do, Taebaek-si, Changjuk-dong, GW, 6.iii.2015) 21 Ditto, head and antenna, macerated 22 Ditto, legs, foreleg to hindleg (left to right), macerated 23 Ditto, genitalia, macerated 24 *Dahlica (Dahlica) somae*, paratype (Korea, Daejeon, Mt. Heungnyongsan, 6.iii.2017) 25 Ditto, head and antenna, macerated 26 Ditto, legs, foreleg to hindleg (left to right), macerated 27 Ditto, genitalia, macerated.

**DNA barcode.** DNA barcode sequences were generated from three individuals. Multiple alignments using the BLAST tool in the NCBI database showed the following species as nearest neighbor: *Dahlica charlottae* with a similarity between 97 and 95%.

**Etymology.** The species is named in honor of Ms. Da-Som Kim, collector of the material.

#### Dahlica (Dahlica) ochrostigma Roh & Byun, sp. n.

http://zoobank.org/EAAEF10F-24B1-4FD8-A7A8-4C9313E4648C Figs 13–19, 20–23, 29, 34, 35

**Type material.** *Holotype.*  $3^{\circ}$  Korea: Gangwon-do, Taebaek-si, Changjuk-dong, 6.iii.2015, S.J. Roh & J.H. Jeon & T.H. Yoo, genitalia mounted on 80% glycerol solution, genitalia No. KNAESJ04, scales of forewing mounted on 80% glycerol solution, scales of forewing No. KNAESSJ03. venation of forewing No. KNAEVSJ03, Deposited at SEL/HNU.

**Paratypes.**  $73^{\circ}$ ,  $39^{\circ}$ . **Korea:**  $33^{\circ}$ ,  $29^{\circ}$ , Gangwon-do, Pyeongchang-gun, Nodong-ri, 6.iii.2015, S.J. Roh & J.H. Jeon & T.H. Yoo, male genitalia mounted in 80% glycerol solution, genitalia No. KNAESJ05, scales of forewing mounted in 80% glycerol solution, scales of forewing No. KNAESSJ04, venation of forewing No. KNAEVSJ04, DNA barcode accession No. of male MF508657, DNA barcode accession No. of female MF664101;  $43^{\circ}$ ,  $19^{\circ}$  Gangwon-do, Taebaek-si, Changjuk-dong, GW, 6.iii.2015, S.J. Roh & J.H. Jeon & T.H. Yoo, scales of forewing mounted in 80% glycerol solution, scales of forewing No. KNAESSJ05, DNAbarcode accession No. of female MF664102. Deposited at SEL/HNU. Other material.  $13^{\circ}$  Korea: Gyeonggi-do, Paju-si, 2.iv.2007, B.W. Lee, genitalia mounted in 80% glycerol solution, genitalia No. KNAESJ06. Deposited at KNAE.

**Diagnosis.** Male, this species is superficially similar to *D. somae* sp. n., but can be distinguished by slightly shorter antennae, a narrow forewing, and the venation of hindwing M3 stalked at 1/4 of M2. This species can be readily differentiated by the dorsum of male genitalia, which is strongly arched to the apical part and in the shape of a hat, and a very short phallus (lateral aspect). Female, apophyses posteriores 1.16 times longer than apophyses anteriores.

**Description.** Adult. Male (Figs 13–19). Wingspan 9.8–11.2 mm (Table 2). Coloration and vestiture: Vertex of head roughly covered with short grayish brown hairs. Thoracic notum covered with brown hairs. Upper side of forewing: ground color gray with sparsely yellow spots; scales (Fig. 16) considerably narrow; apical margin usually produced into two to three laciniations (classes 1–2) (Table 2). Postmarginal part of hindwing present with long shiny white hairs. Structure: head slightly small, compound eyes relatively large; ocelli absent. Antennae filiform (Fig. 15), less than 1/2 forewing. Forewing: short and narrow; costa straight; apex strongly arched to termen, discoidal cell 0.67 times as long as forewing; venation (Fig. 29) with nine veins, originating at the discoidal cell; intercalary cell absent and accessory cell present; Sc reaching to 3/5 costa;



Figures 28–29. Wing venation of males. 28 *Dahlica (Dahlica) somae*, holotype (KNAEVSJ01) 29 *Dahlica (Dahlica) ochrostigma*, holotype (KNAEVSJ03).



**Figures 30–35.** Microhabitat and larval cases with pupal exuviae. **30** Microhabitat of *Dahlica (Dahlica)* ochrostigma (Korea: Gangwon-do, Pyeongchang-gun, Nodong-ri, 6.iii.2015). **31** Ditto, close up **32** Male of *Dahlica (Dahlica) somae*, larval case with pupal exuviae **33** Female of *D. (D.) somae*, ditto **34** Male of *D. (D.) ochrostigma*, ditto **35** female of *D. (D.) ochrostigma*, ditto.



**Figure 36.** A Neighbor-joining tree, generated under the Kimura 2 parameter model (MEGA 6, Tamura et al. 2013) for the species of genera *Dahlica* and *Narycia* (DNA barcode data from NCBI). Branch lengths represent the number of substitutions per site as percentage.

 $R_4$  and  $R_5$  fused;  $R_3$  and  $R_4 + R_5$  originating at apical corner of anterior part of discoidal cell;  $M_1$  and  $M_2$  parallel;  $M_2$  and  $M_3$  stalked at apical corner of posterior part of the discoidal cell;  $Cu_1$  and  $Cu_2$  parallel. Hindwing (Fig. 29): costa straight; discoidal cell 0.52 times as long as hindwing; Sc straight and reaching to 4/5 costa; R originated at apical corner of anterior part of discoidal cell and reaching the apex;  $M_1$  and  $M_2$  parallel,  $M_3$  stalked at 1/4  $M_2$  (Table 2);  $Cu_1$  and  $Cu_2$  parallel. Legs covered with shiny brown scales, epiphysis absent (Fig. 17).

Female (Figs 20-23). 4.5 mm in length. Coloration: Head dark-brown. Meso and metanotum red-brown. Membranous areas of abdomen yellow. Abdomen clothed with light brown scales; corethrogyne densely covered with yellowish white hairs at only ventral part. Structure: Apterous. Head slightly small, antennae relatively developed and long. Legs well developed, slightly long, tarsi 4-segmented.

Male genitalia (Figs 18, 19). In lateral aspect. Genitalia index, 0.79-1.08 (Table 2). Dorsum strongly arched to apical part with the shape of a hat. Saccus relatively short; ampulla slightly long and club shape, setae sparsely; harpe short with needle shape; phallus slender and short with whip shape. In dorso-ventral aspect, uncus slightly concave shape; gnathos and juxta absent; valva slightly narrow and apical part produced into weak rounded claviform.

Female genitalia (Fig. 23). Oviscapt and ostium bursae sclerotized. Apophyses posteriores 1.16 times longer than apophyses anteriores, slender. Sclerotizitions of seventh sternite present with bundle of hairs.

Larval case (Figs 34, 35). Length 3.6-3.9 mm. Their cases are superficially similar to those of *D. somae* sp. n.

#### Distribution. Korea.

DNA barcode. DNA barcode sequences were generated from three individuals (Table 1). Multiple alignments using the BLAST tool in the NCBI database showed the following species as nearest neighbor, Dahlica charlottae with a similarity between 96 and 94%.

**Etymology.** The specific name is derived from the Greek words *ochro* and *stigma* (= pale spots), referring to the forewing pattern.

#### Discussion

The taxonomy of *Dahlica* has until recently been confusing owing to the similar morphology of the species in this genus and those in the allied genera Siederia, Postsolenobia, Brevantennia, and Praesolenobia. The proposal by Arnscheid and Weidlich (2017) to treat all these as subgenera of Dahlica has partly solved this problem. In this study, two new Korean species of *Dahlica* were reported for the first time with COI barcodes (Table 1). The results of comparison with related taxa, including subgenera of Dahlica, revealed no distinct differences (Fig 36). Therefore, the taxonomic positions of the species in genus Dahlica needs to be redefined through future systematic studies with additional samples.

Most species of the genus *Dahlica* have been reported from Europe (48 species) to date, only one species, D. (D.) parthenogenesis Saigusa, 1961 was collected in Japan (Saigusa 1961). Thus, the two new species described in this study represent the first records for continental East Asia and serve as important basic data for future research on this genus and allied taxa in Asia.

#### Acknowledgements

We thank Mr. JH Jeon, TH Yoo, and Ms. DS Kim (Systematic Entomology Lab., Hannam University Korea) for their assistance in collecting specimens. This study was carried out with the support of the 'R&D Program for Forest Science Technology (Project No.: S111616L050120)' provided by Korea Forest Service (Korea Forestry Promotion Institute) and Korea National Arboretum (Project No. KNA1-1-20, 16-1).

#### References

- Arnscheid WR, Weidlich M (2017) Microlepidoptera of Europe; Vol. 8 Psychidae. Brill, Leiden, 423 pp.
- Arnscheid WR (2016) A new species of genus *Dahlica* Enderlein, 1912 form the Pyrenees of Aragon (Province of Huesca) in Spain (Lepidoptera: Psychidae: Dahlicini). SHILAP Revista de Lepidopterologia 44(173): 39–43.
- Davis DR, Robinson GS (1998) The Tineoidea and Gracillarioidea. In: Kristensen NP (Ed.) Lepidoptera, Moths and Butterflies. Vol 1: Evolution, Systematics and Biogeography. Handbook of Zoology 4. Walter de Gruyter, Berlin and New York, 91–117. https://doi. org/10.1515/9783110804744.91
- Dierl W (1964) Cytologie, Morphologie und Anatomie der Sackspinner *Fumea casta* (Pallas) und crassionella (Bruand) sowie *Bruandia comitella* (Bruand) (Lepidoptera, Psychidae) mit Kreuzungsversuchen zur Klärung der Artspezifität. Zoologische Jahrbucher Systematik 91: 201–270.
- Elzinga JA, Jokela J, Shama LNS (2013) Large variation in mitochondrial DNA of sexual and parthenogenetic *Dahlica triquetrella* (Lepidoptera: Psychidae) shows multiple origins of parthenogenesis. BMC Evolutionary Biology 13: 90. https://doi.org/10.1186/1471-2148-13-90
- Enderlein G (1912) I. Wissenschaftliche Mitteilungen, 2. Zur Kenntnis der Zygophtalmen. Zoologischer Anzeiger 40: 261–282.
- Grapputo A, Kumpulainen T, Mappes J (2005) Phylogeny and evolution of parthenogenesis in Finnish bagworm moth species (Lepidoptera: Psychidae: Naryciinae) based on mtDNAmarker. Annales Zoologici Fennici 42:141–160.
- Hebert DN, Penton EH, Burns JM, Janzen DH, Hallwachs W (2004) Ten species in one: DNA barcoding reveals cryptic species in the neotropical skipper butterfly Astraptes fulgerator. Proceedings of the National Academy of Sciences of the United States of America 101: 14812–14817. https://doi.org/10.1073/pnas.0406166101
- Herrmann R (1988) *Dahlica marmorella* sp. n. eine neue Psychidae aus Italien (Lepidoptera: Psychidae). Nota Lepidopterologica 10(4): 203–208.
- Herrmann R, Weidlich M (1999) Psychidenbeobachtungen in Westrumänien.-Teil 2. Beschreibung von Siederia transsylvanica sp. n. (Psychidae). Nota Lepidopterologica 22(1): 10–16.
- Hübner J (1796-1836) Sammlung europäischer Schmetterlinge. 8. Horde. Die Schaben; nach der Natur geordnet, beschrieben und vorgestellt. Augsburg, 1–78, pls 1–71.
- Hori H (1926) A new Psychid from Japan. Kontyû 1: 28-30.

- Katoh K, Toh H (2008) Recent developments in the MAFFT multiple sequence alignment program. Briefings in Bioinformatics 9(4): 286–298. https://doi.org/10.1093/bib/bbn013
- Kristensen NP (2003) Skeleton and muscles: adults. In: Kristensen NP (Ed.) Lepidoptera, Moths and Butterflies, 2. Morphology, physiology and development. De Gruyter, Berlin, New York. Handbuch der Zoologie/ Handbook of Zoology 4(36): 39–131. https://doi. org/10.1515/9783110893724.39
- Kumpulainen T (2004) The evolution and maintenance of reproductive strategies in bag worm moth (Lepidoptera: Psychidae). Jyväskylä Studies in Biological and Environmental Sciences 132: 1–42.
- Kumpulainen T, Grapputo A, Mappes J (2004) Parasites and sexual reproduction in psychid moths. Evolution 58: 1511–1520. https://doi.org/10.1111/j.0014-3820.2004.tb01731.x
- Meier H (1958) Der taxonomische Wert der Hinterflügel-Aderung bei den Gattungen Brevantennia Sieder und Solenobia Duponchel (Lep., Psych.). Mitteilungen des naturwissenschaftlichen Vereins für Steiermark 88: 178–192.
- Meyrick E (1935) Exotic Microlepidoptera, Vol. 4. Taylor and Francis, London, 577-608.
- Mutanen M, Wahlberg N, Kaila L (2010) Comprehensive gene and taxon coverage elucidates radiation patterns in moths and bufferflies. Proceedings of the Royal Society B Biological Sciences 277: 2839–2848. https://doi.org/10.1098/rspb.2010.0392
- Nieukerken EJ van, Kaila L, Kitching IJ, Kristensen NP, Lees DC, Minet J, Mitter C, Mutanen M, Regier JC, Simonsen TJ, Wahlberg N, Yen SH, Zahiri R, Adamski D, Baixeras J, Bartsch D, Bengtsson BA, Brown JW, Bucheli SR, Davis DR, De Prins J, De Prins W, Epstein ME, Gentili-Poole P, Gielis C, Hattenschwiler P, Hausmann A, Holloway JD, Kallies A, Karsholt O, Kawahara AY, Koster SJC, Kozlov MV, Lafontaine JD, Lamas G, Landry JF, Lee S, Nuss M, Park KT, Penz C, Rota J, Schitlmeister A, Schmidt BC, Sohn JC, Solis MA, Tarmann GM, Warren AD, Weller S, Yakovlev RV, Zolotuhin VV, Zwick A (2011) Order Lepidoptera Linnaeus, 1758. In: Zhang ZQ (Ed.) Animal biodiversity: an outline of higher-level classification and survey of taxonomic richness. Zootaxa 3148: 212–221.
- Rekelj J, Predovnik Ž (2014) *Dahlica goltella* sp. n., a new bagworm species from Slovenia (Lepidoptera: Psychidae). Acta Entomologica Slovenica 22(1): 5–18.
- Regier JC, Mitter C, Davis DR, Harrison TL, Sohn JC, Cummings MP, Zwick A, Mitter KT (2015) A molecular phylogeny and revised classification for the oldest ditrysian moth lineages (Lepidoptera: Tineoidea), with implications for ancestral feeding habits of the mega-diverse Ditrysia. Systematic Entomology 40: 409–432. https://doi.org/10.1111/syen.12110
- Roh SJ, Byun BK (2017a) First discovery of the Lichen-Feeding Moth *Bacotia sakabei* (Lepidoptera: Psychidae) from Korea. Animal Systematics, Evolution and Diversity 33(1): 60–64. https://doi.org/10.5635/ASED.2017.33.1.064
- Roh SJ, Byun BK (2017b) Two species of the subfamily Psychinae (Lepidoptera: Psychidae) new to Korea. Journal of Asia-Pacific Biodiversity 10(2): 224–227. https://doi.org/10.1016/j. japb.2017.04.014
- Roh SJ, Banasiak G, Byun BK (2016) A new and an unrecorded species of the family Psychidae (Lepidoptera) from Korea, with an annotated catalogue. Journal of Natural History 50(11/12): 669–680. https://doi.org/10.1080/00222933.2015.1082654

- Roh SJ, Byun BK (2016) Discovery of *Ceratosticha leptodeta* Meyrick (Lepidoptera: Psychidae) from Korea. Journal of Asia-Pacific Biodiversity 9(1): 91–93. https://doi.org/10.1016/j. japb.2015.12.009
- Saigusa T (1961) Systematic studies of *Diplodoma* and its allied genera in Japan. Sieboldia. II/4: 261–315.
- Saigusa T, Sugimoto M (2014) Japanese species of the genus *Proutia* Tutt, 1899 (Lepidoptera: Psychidae). Zootaxa 3869: 143–152. https://doi.org/10.11646/zootaxa.3869.2.3
- Sauter W (1956) Morphologie und Systematik der schweizerischen *Solenobia-* Arten. Revue Suisse de Zoologie 63: 451–550. https://doi.org/10.5962/bhl.part.75469
- Sauter W, Hättenschwiler P (1991) Zum System der palaearktischen Psychiden (Psychidae) 1 Teil: Liste der paläarktischen Arten. Nota lepidopterologica 22: 262–295.
- Sauter W, Hättenschwiler P (1999) Zum System der palaearktischen Psychiden (Psychidae) 2 Teil: Bestimmungsschlüssel für die Gattungen. Nota lepidopterologica 22: 262–295.
- Seino A (1981) A new psychid species of *Bacotia* from Japan (Lepidoptera). Tyô to Ga 31: 121–125.
- Sieder L (1953) Vorerbeit zu einer Monographie über sie Gattung Solenobia Z. (Lepidopt. Psychidae-Taleporiinae). Zeitschrift der Wiener Entomologischen Gesellschaft 38(5): 113–128.
- Sobczyk T (2011) World catalogue of insects; Vol.10 Psychidae(Lepidoptera). Apollo Books, Stenstrup, 467 pp.
- Sugimoto M (2009a) A comparative study of larval cases of Japanese Psychidae(Lepidoptera). Japanese Journal of Entomology (NS) 12: 1–15.
- Sugimoto M (2009b) A comparative study of larval cases of Japanese Psychidae(Lepidoptera). Japanese Journal of Entomology (NS) 12: 17–29.
- Suomalainen E (1980) The Solenobiinae species of Finland (Lepidoptera: Psychidae), with description of a new species. Entomologica Scandinavica 11: 458–466. https://doi.org/10.1163/187631280794710042
- Tamura K, Stecher G, Peterson D, Filipski A, Kumar S (2013) MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. Molecular Biology and Evolution 30: 2725–2729. https://doi.org/10.1093/molbev/mst197

ZooKeys 733: 65–97 (2018) doi: 10.3897/zookeys.733.20159 http://zookeys.pensoft.net

CHECKLIST



# Fishes of the Cusiana River (Meta River basin, Colombia), with an identification key to its species

Alexander Urbano-Bonilla<sup>1</sup>, Gustavo A. Ballen<sup>1,2</sup>, Guido A. Herrera-R<sup>1,3</sup>, Jhon Zamudio<sup>1,4</sup>, Edgar E. Herrera-Collazos<sup>1</sup>, Carlos DoNascimiento<sup>5</sup>, Saúl Prada-Pedreros<sup>1</sup>, Javier A. Maldonado-Ocampo<sup>1</sup>

Laboratorio de Ictiología, Unidad de Ecología y Sistemática (UNESIS), Departamento de Biología, Facultad de Ciencias, Pontificia Universidad Javeriana, Carrera 7 N° 43-82, Bogotá, D.C., Colombia 2 Museu de Zoologia da Universidade de São Paulo, Caixa Postal 42494, 04218-970 São Paulo, SP, Brazil 3 Université Paul Sabatier, 118 Route de Narbonne, 31062, Toulouse, France 4 Grupo de Investigaciones territoriales para el uso y conservación de la Biodiversidad, Fundación Reserva Natural La Palmita - Centro de Investigación, Carrera 4 N° 58-59, Oficina 301, Bogotá, D.C., Colombia 5 Colecciones del Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, Claustro de San Agustín, Carrera 8 N° 15-08. Villa de Leyva, Colombia

Corresponding author: *Alexander Urbano-Bonilla* (bio.ictiologia@gmail.com), *Javier A. Maldonado-Ocampo* (maldonadoj@javeriana.edu.co)

Academic editor: D. Bloom   Received 9 August	2017   Accepted 2 January 2018   Published 26 January 2018
http://zoobank.org/6B80,	

**Citation:** Urbano-Bonilla A, Ballen GA, Herrera-R GA, Zamudio J, Herrera-Collazos EE, DoNascimiento C, Prada-Pedreros S, Maldonado-Ocampo JA (2018) Fishes of the Cusiana River (Meta River basin, Colombia), with an identification key to its species. ZooKeys 733: 65–97. https://doi.org/10.3897/zookeys.733.20159

#### Abstract

The Cusiana River sub-basin has been identified as a priority conservation area in the Orinoco region in Colombia due to its high species diversity. This study presents an updated checklist and identification key for fishes of the Cusiana River sub-basin. The checklist was assembled through direct examination of specimens deposited in the main Colombian ichthyological collections. A total of 2020 lots from 167 different localities from the Cusiana River sub-basin were examined and ranged from 153 to 2970 m in elevation. The highest number of records were from the piedmont region (1091, 54.0 %), followed by the Llanos (878, 43.5 %) and Andean (51, 2.5 %). 241 species distributed in 9 orders, 40 families, and 158 genera were found. The fish species richness observed (241), represents 77.7 % of the 314 estimated species (95 % CI=276.1–394.8). The use of databases to develop lists of fish species is not entirely reliable; therefore taxonomic verification of specimens in collections is essential. The results will facilitate comparisons with other sub-basins of the Orinoquia, which are not categorized as areas of importance for conservation in Colombia.

Copyright Alexander Urbano-Bonilla et al. This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

#### Resumen

La sub-cuenca del rio Cusiana ha sido designada como una de las áreas prioritarias para la conservación en la región del Orinoco en Colombia debido a su alta diversidad de especies. Este estudio presenta una lista actualizada y una clave de identificación para los peces del área. Para ello se revisaron los especímenes depositados en las principales colecciones ictiológicas colombianas. Se examinaron un total de 2020 lotes de 167 localidades diferentes de la sub-cuenca del río Cusiana, que oscilaron entre 153 y 2970 m en altitud. El mayor número de registros corresponde a la región del piedemonte (1091, 54.0%), seguida por los Llanos (878, 43.5%) y Andina (51, 2.5%). Encontramos 241 especies distribuidas en 9 órdenes, 40 familias y 158 géneros. La riqueza de especies de peces observada (241), representan el 77.7% de las 314 especies estimadas (IC 95% = 276.1-394.8). El uso de bases de datos para elaborar listas de especies de peces no es del todo fiable, por lo tanto, es esencial la verificación taxonómica de los especímenes en las colecciones. Estos resultados permitirán evaluar otras sub-cuencas de la Orinoquia, las cuales no están categorizadas como áreas de importancia para la conservación en Colombia.

#### **Keywords**

Andean Orinoquia, Casanare, conservation, diversity, Llanos, Piedmont, species richness

#### **Palabras clave**

Diversidad, Casanare, Conservación, Llanos, Orinoquia Andina, Piedemonte, Riqueza de especies

#### Introduction

The Orinoco River, with an estimated richness of 1002 species of freshwater fishes, is the second most diverse drainage in the Neotropical region (Reis et al. 2016). Nonetheless, the basin has been exposed to increasing threats due to human activities that place the enormous fish diversity at risk (Barletta et al. 2010, Rybicki and Hanski 2013, Lasso et al. 2016). The systems draining the Andean region (western tributaries of the Orinoco) are considered the most threatened at basin scale (Rodríguez et al. 2007, Machado-Allison et al. 2010, Lasso et al. 2016). The rivers originating in the Andes are heavily exposed to threats like habitat fragmentation, contamination, deforestation, the introduction of non-native species and mining (Machado-Allison et al. 2010, Anderson and Maldonado-Ocampo 2011, Lasso et al. 2016). Additionally, large gaps regarding the basic knowledge of fish diversity of the Andean sub-basins are persistent, especially in Colombia (Maldonado-Ocampo et al. 2008, Machado-Allison et al. 2010, Lasso et al. 2016). Filling those gaps are essential to guide adequate conservation efforts for the freshwater ecosystems and therefore face the threats above mentioned.

The Meta River basin, with headwaters on the Eastern Cordillera of Colombia, is one of the major tributaries of the Orinoco River (Usma-Oviedo et al. 2016). Studies on its fish diversity (e.g., Urbano-Bonilla et al. 2009, 2014, Villa-Navarro et al. 2011, Maldonado-Ocampo et al. 2013, Urbano-Bonilla and Maldonado-Ocampo 2013), and recent efforts (Zamudio et al. 2008, Urbano-Bonilla et al. 2016) have advanced our understanding of the ecology of some species. The Cusiana River sub-basin is one of the best-known Andean tributaries of the Meta River basin; the first inventories of its fish diversity dated back to the 90's with the establishment of oil companies in the area. The Cusiana River sub-basin has been considered as a conservation priority area for the Orinoco biodiversity due to its high diversity in several groups (Lasso et al. 2010, Trujillo et al. 2011), including fishes (Villa-Navarro et al. 2011).

Here an updated checklist and an identification key are presented for the fishes of the Cusiana River sub-basin. We hope our results may establish a guideline that can be replicated in other basins of the Orinoco drainage.

#### Materials and methods

The Cusiana River sub-basin has an extension of 7324 km<sup>2</sup> and 271 km in length, originating at 3800 m asl on the eastern slope of the Eastern Cordillera in the Quebradas La Iglesia, Melgarejo, and Las Cañas, Boyacá Department (5°35'N, 72°47'W), and empties at 150 m asl in the Meta river, Casanare Department (4°31'N, 71°51'W) (IGAC 1999) (Fig. 1). The Cusiana River sub-basin was divided by altitudinal limits in three distinctive regions based on Abell et al. (2008): Llanos (139–300 m asl), Pied-mont (300–1235 m asl) and High Andes (1235–3000 m asl).

The checklist was assembled by examining specimens deposited in Colombian ichthyological collections. Acronyms used in the text follow Sabaj-Pérez (2016) except uncatalogued material housed at Fundación Universitaria del Trópico Americano (UNITROPICO). The taxonomic list follows the classification system proposed by Reis et al. (2003) with recent modifications proposed by Oliveira et al. (2011) for characiform families, Betancur-R et al. (2016) at high-level groups for osteichthyans in general, and Thomaz et al. (2015) for genera of the Stevardiinae. Valid species names were confirmed through queries on the Catalog of Fishes of the California Academy of Sciences (Eschmeyer et al. 2017). Species were categorized as endemic (DoNascimiento et al. 2017), threatened (Mojica et al. 2012), migratory (Usma-Oviedo et al. 2013), and species subject of conservation (González et al. 2015).

Species richness interpolation and extrapolation was calculated following Chao et al. (2014) and using the package iNEXT 2.0.12 (Hsieh et al. 2016) for R v.3.4.0 (R Core Development Team 2017). The number of localities were obtained per Orinoco basin from the data set of the "Catalogue of the Freshwater Fishes of Colombia" (Do-Nascimiento et al. 2017).

To construct the key (for order, families and species), original descriptions of species, taxonomic revisions, and direct examination of specimens were used. Finally, in order to share the information produced herein, the dataset was uploaded to SiB Colombia's data portal (GBIF Colombia Node) in accordance with their protocols for species lists. For the latter, the complete dataset was structured and standardized to comply with the international biodiversity standard: Darwin Core standard (Wieczorek et al. 2012). After mounting the dataset on a Darwin Core spreadsheet template, it was uploaded to SiB Colombia's Integrated Publishing Tool for international visualization in their data portal. A DOI was provided by SiB Colombia for the shared dataset available at http://doi.org/10.15472/er3svl, all the results, discussion and quantities herein cited follow the version 1.8 of the published dataset.



Figure 1. Collection localities in the Cusiana River sub-basin.

#### Results

In total, 2020 lots from 167 different localities from the Cusiana River sub-basin ranging from 152 to 2970 m asl were examined. Most of the records were found in the piedmont (1091, 54.0%), followed by the llanos (878, 43.5%) and Andean Orinoquia (51, 2.5%), suggesting sampling bias in elevation for this drainage, being inverselyproportional to elevation (Fig. 2). The number of localities in Cusiana River sub-basin represents the quantile 0.83 among the tributaries of the Orinoco drainage in Colombia (Table 1). 241 species were found distributed in nine orders, 40 families, and 158 genera. The order Characiformes showed the highest richness with 106 species, followed by Siluriformes (89), Gymnotiformes (20), and Cichliformes (15). The remaining orders were represented by one to three species. The most speciose families were the Characidae (54), Loricariidae (30), Cichlidae (15), Heptapteridae (15), Pimelodidae (11), and Curimatidae (9), while the 34 remaining families were represented by 1 - 8 species. Extrapolation suggests that the expected richness for the Cusiana River subbasin is roughly 314 species (95% CI = 276.1-394.8) (Fig. 3); with the observed richness corresponding to around 77.7% the expected richness.

Concerning threatened species, five are currently categorized as Vulnerable (Potamotrygon motoro, Brachyplatystoma vaillantii, Pseudoplatystoma metaense, P. orinocoense,

#### Distribution of sampling records along the elevational gradient



**Figure 2.** Distribution of sampling records along the Elevational gradient in the Cusiana River subbasin. Asterisks indicate categories with the lowest sampling along the elevational gradient.

Basin	Number of sampling locations
Guamal-Humadea	213
Guacavía-Guatiquía-Humea	174
Cusiana	167
Ariari	74
Upía	61
Guayuriba	57
Cravo-Norte	56
Pauto	56
Cravo-Sur	53
Arauca	43
Guayabero	35
Tua	29

Table 1. Number of sampling sites per basin.

and Zungaro zungaro), and two as Nearly Threatened (*Potamotrygon orbignyi* and *Sorubim lima*). There are 34 species endemic to the Orinoco drainage, 20 are migratory, and 8 are subjects of conservation. A total of 50 species are new records for the Cusiana River sub-basin, while *Cetopsorhamdia shermani* and *Rhamdia muelleri* are also new records for Colombia. Some species from the genera *Andinoacara, Astroblepus, Ceratobranchia, Cetopsorhamdia, Chaetostoma, Characidium, Corydoras, Creagrutus, Curimatopsis, Hypostomus, Imparfinis, Microglanis, Parodon, Parotocinclus, Pimelodella, Poecilia, Spatuloricaria*, and *Trichomycterus*, require further revision by specialists.



**Figure 3.** Species accumulation curve in the Cusiana River sub-basin. Abbreviations: S.obs = observed richness, S.est = estimated richness. Continuous line represents interpolation, and dashed line represents extrapolation.

# Key to orders, families, and species of fishes of the Cusiana River sub-basin

1	Five pairs of gills in ventral position
_	Two lateral gill openings, or just one gill opening under head2
2	Eyes located on the same side of body
_	Eyes located on opposite sides of body
3	Dorsal fin absent
_	Dorsal fin present
4	Anal fin absent; one ventral gill opening
	SYNBRANCHIFORMES: Synbranchidae: Synbranchus marmoratus
_	Anal fin present, always long, two lateral gill openings
	GYMNOTIFORMES (20 species)
5	Body naked or covered with bony plates; barbels present near the mouth
	SILURIFORMES (90 species)
_	Body totally or partially covered with scales; barbels absent

6	Dorsal and anal fins with two or more spines; scales ctenoid7
_	Dorsal and anal fins without spines; scales cycloid (ctenoid in some groups)8
7	Lateral line interrupted CICHLIFORMES (15 species)
_	Lateral line continuous
	ACANTHURIFORMES: Sciaenidae (2 species)
8	Mouth superior and somewhat protractile; adipose fin absent
	CYPRINODONTIFORMES (3 species)
_	Mouth usually in terminal position, never protractile; adipose fin usually pre-
	sent CHARACIFORMES (106 species)

## MYLIOBATIFORMES Potamotrygonidae

# PLEURONECTIFORMES Achiridae

1	Pectoral fins present; gill openings wide and extending ventrally on both sides
	of head
_	Pectoral fins absent; gill openings limited to a narrow slit and never con-
	nected ventrally to both sides of head Apionichthys saula

# **GYMNOTIFORMES**

1	Caudal fin and dorsal filament present	Apteronotidae
_	Caudal fin and dorsal filament absent	2
2	Mouth in upper position, body cylindrical	Gymnotidae
_	Mouth terminal or subterminal, body compressed	
3	Teeth present; nares tubular	Sternopygidae
_	Teeth absent; anterior nares not tubular	
4	Absence of mental accessory electric organ	•••••
	Hypopomidae: Brachyhypo	opomus brevirostris
_	Presence of mental accessory electric organ	Rhamphichthyidae

# Apteronotidae

1	Lower jaw with a distinct V-shaped median groove accommodating the
	pointed decurved upper jaw
_	Lower jaw without a V-shaped median groove
2	Snout tubular
_	Snout obtuse or elongate but not tubular
3	Absence of teeth on upper jawPlatyurosternarchus macrostoma
_	Presence of teeth on upper jaw
4	Total anal-fin rays 212–242 Sternarchorbynchus oxyrbynchus
_	Total anal-fin rays 193–210 Sternarchorhynchus roseni
5	Mid-dorsal pale stripe absent
_	Mid-dorsal pale stripe present
6	Presence of two clear bands surrounding caudal peduncle Apteronotus albifrons
_	Presence of a single clear band surrounding base of caudal peduncle7
7	More than 10 scales above lateral line Apteronotus galvisi
_	10 or fewer scales above lateral line Apteronotus bonapartii

# Gymnotidae

1	Without a particular color pattern on body; anal fin confluent with tail
	Electrophorus electricus
_	Body color pattern formed by dark oblique bands alternating with pale bands;
	anal fin not confluent with tail
2	Obliquely-oriented dark bands or band pairs with straight or high-contrast
	margins, 23 dark bands (usually more than 30) Gymnotus cataniapo
_	Obliquely-oriented dark bands or band pairs with irregular and wavy margins
	23 Gymnotus carapo septentrionalis

# Sternopygidae

1	Orbital margin free; all anal-fin rays simple; background color variable from uniformly gray to black; humeral black blotch present, rarely diffused; white narrow band under the lateral line, from the midbody to end of anal fin
	Sternopygus macrurus
_	Orbital margin continuous; background color variable from translucent to
	white; humeral blotch absent
2	Anal fin either completely black or with a black margin; no horizontal dark
	stripes on body; scales above lateral line 15-18 Eigenmannia limbata
_	Anal fin hyaline; 2 or 3 horizontal dark stripes on body; scales above lateral
	line 8–11Eigenmannia sp.
## Rhamphichthyidae

1	Anterior nares inside upper lip
_	Anterior nares outside upper lip
2	Absence of sixth infraorbital bone
_	Presence of sixth infraorbital bone, as a narrow tube, positioned vertically,
	parallel to posterior border of eye
3	Number of pectoral-fin rays fewer than 16
	Gymnorhamphichthys hypostomus
_	Number of pectoral-fin rays more than 164
4	Anal fin usually clear or hyaline
_	Anal fin usually dark with a terminal dark band Rhamphichthys rostratus

### **CYPRINODONTIFORMES**

1	Pectoral fin with 1–2 unbranched rays	.Poeciliidae: Poecilia sp.
_	Pectoral fin with all rays branched	Cynolebiidae

## Cynolebiidae

1	Scales on the ventral surface of the head	Anablepsoides sp.
_	Scales absent on the ventral surface of the head	Rachovia maculipinnis

### **SILURIFORMES**

1	Mental barbels absent; ventral mouth in the form of a sucking disk with wide
	lower lip2
_	Mental barbels present; terminal mouth
2	Body naked Astroblepidae
_	Body covered with bony plates Loricariidae
3	Body depressed; skin covered with tubercles and completely keratinized
	Aspredinidae
_	Body not depressed; skin without tubercles
4	Body covered with bony plates
_	Body not covered with bony plates
5	Sides of body covered with two rows of bony plates
_	Sides of body with a mid-lateral row of dermal plates armed with a lateral
	thornDoradidae
6	Opercle and interopercle with odontodes Trichomycteridae
_	Opercle and interopercle without odontodes7

7	Lateral-line system branched on head	Pimelodidae
_	Lateral-line system simple, not branched on head	8
8	Suborbital sulcus present	Auchenipteridae
_	Suborbital sulcus absent	9
9	Adipose fin absent	Cetopsidae
_	Adipose fin present	
10	Eyes set on anteriormost half of head, thick skin	Pseudopimelodidae
_	Eyes set on posteriormost half of head, thin skin	Heptapteridae

## Astroblepidae

1	Adipose fin present	Astrobletus sp.
_	Adipose fin absent	Astroblepus mariae

## Loricariidae

1	Tail strongly depressed
_	Tail oval in cross-section
2	Dorsal-fin insertion in front of the anal-fin insertion
_	Dorsal-fin insertion anterior to the anal-fin insertion
3	Three rows of abdominal plates <i>Farlowella mariaelenae</i>
_	Two rows of abdominal plates4
4	Breeding odontodes on preorbital ridge present Farlowella acus
_	Breeding odontodes on preorbital ridge absent Farlowella vittata
5	Teeth villiform
_	Teeth spoon-shaped, elongate, comb-shaped or even absent but never vil-
	liform7
6	Pectoral-fin rays i,7
_	Pectoral-fin rays i,6 Sturisoma tenuirostre
7	Premaxillary teeth larger than dentary teeth Loricaria cataphracta
_	Premaxillary teeth equal to or smaller than dentary teeth
8	Snout round in outline; upper lip with long filaments covering mouth open-
	ingDentectus barbarmatus
_	Snout acute to ovoid in outline; filaments covering mouth opening absent9
9	Dentary teeth larger than premaxillary teeth Rineloricaria eigenmanni
_	Dentary teeth smaller than or equal to premaxillary teeth10
10	A pair of digitiform papillae on rictal region of mouth and an additional pair
	on mouth roof
_	Digitiform papillae on rictal region of mouth but absent on its mouth roof.

11	Anterior margin of abdominal plate cover oval in outline; fringes on upper lip darkly pigmented
_	Anterior margin of abdominal plate cover irregular in outline; fringes on up-
	per lip unpigmented
12	Snout naked, not covered with plates
_	Snout covered with plates 17
13	Eleshy tentacles on shout present: three series of lateral plate series on caudal
15	peduncle
	Electricite and an ansatt character free parties of lateral plate series on enadel
_	Fleshy tentacies on shout absent; nye series of fateral plate series on caudal
1/	cl l l l l l l l l l l l l l l l l l l
14	Cheek odontodes straight; fleshy excrescence on parieto-supraoccipital ab-
	sent Chaetostoma dorsale
-	Cheek odontodes curved; fleshy excrescence on parieto-supraoccipital pre- sent
15	No dark spots on any fin but dark stripes present along rays in all fins; en-
	larged second unbranched ray in anal fin bearing two fleshy ridges in mature males
_	Dark spots on fins; anal fin never bearing paired fleshy ridges in mature
	males
16	Ventral portion of body posterior to pelvic-fin insertion light, never covered with
10	abundant dark spots: pectoral-fin spine with dark spots <b>Chartostoma sp</b>
_	Body uniformly spotted: pectoral-fin spine without spots
17	Posterior serre on pectoral fin spine present <b>Hubattationa machadai</b>
1 /	Posterior serve on pectoral fin spine present
10	rostenioi seriae on pectorai-nii spine absent
18	Spoon-snaped teetn
_	Villiform teeth
19	Cheek odontodes absent Hypostomus plecostomoides
-	Cheek odontodes present and erectile20
20	Dark background with yellow or white vertical and irregular bands; small adult size
_	Color pattern consisting of horizontal stripes: large adult size
	Panaque niorolineatus
21	Eves visible in ventral view: adipose fin absent 22
_	Eyes not visible in ventral view: adipose fin present 23
-	Twenty six or more promovillary teeth 21 or more mondibular teeth, teeth
	I wenty-six of more premaxinary teeth, 21 of more mandoular teeth; teeth
	siender, tightly spaced along the jaw rami, spacing between teeth equal to or
	greater than the tooth shaft width, tooth cusps small <b>Otocinclus huaorani</b>
_	Fewer than 25 premaxillary teeth, 20 mandibular teeth; teeth robust, spacing
	between teeth equal to or greater than tooth shaft width, tooth cusps robust
	Otocinclus vittatus
23	Coracoid bearing odontodes in ventral surfaceParotocinclus sp.

24	Setiform odontodes on sides of head; three series of lateral plates on caudal
_	No setiform odontodes on sides of head; five series of lateral plates on caudal
25	Hypertrophied cheek odontodes surpassing base of pectoral fin; pectoral-fin spine reaching or surpassing tip of pelvic-fin leading ray
	Dolichancistrus fuesslii
_	No hypertrophied cheek odontodes; pectoral-fin spine not reaching tip of pelvic-fin leading ray
26	Multiple buccal papillae
_	Single medial buccal papilla
27	More than 11 branched dorsal-fin rays Pterygoplichthys multiradiatus
_	Less than 11 branched dorsal-fin rays
28	Lower caudal-fin lobe dark and upper lobe light, no spots on caudal-fin; cau-
	dal peduncle elongated Aphanotorulus emarginatus
_	Spots on caudal fin, no distinct dark background on lower caudal-fin lobe; caudal peduncle not elongated
29	Dark blotches closely-set on dorsal fin
_	Dark spots arranged in two longitudinal series on interradial membranes, no
	blotches on dorsal fin
	ferences in coloration are preliminary observations based on limited samples along with type specimens, further and more extensive sampling will be re- quired in order to test such differences between <i>Hypostomus argus</i> and <i>H. nice-</i> <i>foroi</i> in the Orinoco basin of Colombia; G.A. Ballen & S. Reinales, in prep.)

## Aspredinidae

1	Dorsal, ventral and lateral series of bony plates present on body; mouth infe-
	rior; hard pectoral spine, without serrae on the anterior margin
_	No obvious bony plates on body; mouth terminal; hard pectoral spine serrate
	on anterior and posterior marginsBunocephalus aloikae

## Callichthyidae

1	Snout depressed; maxillary barbel long, usually extending beyond gill open-
	ing
_	Snout compressed; maxillary barbel short, usually not extending beyond eye 3
2	Caudal fin forked, without conspicuous dark bands; dorsal fin spine about
	half the length of the first branched rays; six branched anal-fin rays

76

k uistai illaigili,
or five branched
Megalechis picta
Corydoras sp.
4
scutes to caudal-
n
rydoras axelrodi
nt of dorsal fin to he caudal pedun- <i>s septentrionalis</i>

## Doradidae

1	Maxillary barbel branched	Leptodoras nelsoni
_	Maxillary barbel simple, not ramified	
2	Anterior and posterior dorsal-fin serrae absent	Amblydoras affinis
_	Anterior, posterior, or both dorsal-fin serrae present	3
3	Procurrent caudal-fin rays modified in fulcra; dark bac	kground with clear
	longitudinal stripePla	itydoras armatulus
_	Procurrent caudal-fin rays not modified in fulcra; color	r pattern without a
	clear longitudinal stripe	4
4	Thick lips, premaxillary and dentary teeth absent	Oxydoras sifontesi
-	Thin lips, premaxillary and dentary teeth present	5
5	Small body, caudal truncate	ydoras bolivarensis
_	Large body, caudal emarginate with two distinct lobes	Pterodoras rivasi

## Trichomycteridae

1	Nasal barbel present (associated with anterior nostril), mouth subterminal2
_	Nasal barbel absent, mouth ventral
2	Sides of body with a dark band or row of spots from just above the gill-opening
	to the base of the upper caudal-fin rays Trichomycterus dorsostriatus
_	Sides of body uniformly pigmented
3	Eight branched pectoral-fin rays (rarely nine rays); a single median epiphyseal
	pore (rarely two asymmetrical pores)
_	Seven branched pectoral-fin rays (rarely six rays); a pair of symmetrical epi-
	physeal pores
4	Lower jaw rami not articulated medially, only joined by soft tissue
	Vandellia beccarii
_	Lower jaw rami medially articulated
	· ·

### Cetopsidae

78

### Heptapteridae

1	Eye margin free, first dorsal and pectoral-fin rays with a pungent spine2
_	Eye margin not free, first dorsal and pectoral-fin rays soft and segmented (at
	least their distal part)
2	Occipital process in contact with the nuchal plate, posterior cranial fontanelle
	open
_	Occipital process not reaching posteriorly the nuchal plate, posterior cranial
	fontanelle closed or reduced
3	Adipose-fin equals to one third of SL or shorter Pimelodella metae
_	Adipose-fin longer than one third of SL
4	Upper caudal-fin lobe narrower and shorter than the lower caudal-fin lobe
	Pimelodella cristata
_	Upper caudal-fin lobe about same width than lower lobe and distinctively
	longer
5	Upper caudal-fin lobe distinctively shorter than the upper lobe: if undam-
2	aged, maxillary barbel surpassing the caudal peduncle <b>Rhamdia muelleri</b>
_	Caudal-fin lobes subequal, maxillary barbel not surpassing the caudal pedun-
	cle Rhamdia avelen
6	Lower caudal-fin lobe longer than upper lobe <b><i>Phenacorhandia taphorni</i></b>
0	Lower caudal fin lobe equal or shorter than upper lobe
7	Lateral line canal incomplete extending only to doreal fin base
/	Nomenno d'anio mania i
	Lateral line canal complete extending to caudal for base
-	Lateral-line canal complete, extending to caudal-lin base

8	Lateral-line canal posteriorly interrupted as a series of short canal segments	
0	Imparfinis microps	
8	Lateral line continuous	
9	Adipose-nn rectangular	
-	Adipose-nn triangular	
10	one fourth of SL, six pale bars across dorsum and sides of body	
	Anal fin origin anterior to adinose fin origin adinose fin shorter than one	
_	fourth of SL, four pale bars across dorsum and sides of body	
	Cetopsorhamdia orinoco	
11	First dorsal and pectoral-fin ray extended as a long filament, maxillary barbel surpassing anal-fin base <i>Imparfinis pseudonemacheir</i>	
_	First dorsal and pectoral-fin ray not extended as a long filament, maxillary	
	barbel not reaching anal-fin origin12	
12	Blotched pattern of body pigmentation13	
_	Uniform dark pattern of pigmentation14	
13	Pelvic-fin origin approximately at vertical through dorsal-fin origin, adipose- fin base extending far posteriorly beyond distal end of anal-fin	
_	Pelvic-fin origin slightly behind mid-length of dorsal-fin base, adipose-fin base	
	ending around same level of distal end of anal fin Cetopsorhamdia shermani	
14	Snout conical and conspicuously projected beyond mouth opening, maxil- lary barbel not surpassing distal end of pectoral fin, dorsal and ventral mar-	
	gins of end of caudal peduncle with a conspicuous white area, caudal fin clear	
	contrasting with dark caudal fin base Cetopsorhamdia aff. picklei	
14	Snout depressed, not projected conspicuously beyond mouth opening, maxil-	
	lary barbel surpassing distal end of pectoral fin, dorsal and ventral margins	
	of end of caudal peduncle dark, pigmented as remaining of body, caudal fin	
	dusky, not contrasting with caudal fin base Imparfinis sp.	

## Pseudopimelodidae

1	Lateral line incomplete; premaxillary tooth patch without posterolateral pro-
	jection; small size
_	Lateral line complete; premaxillary tooth patch with posterolateral projec-
	tion; large size
2	Bar-shaped blotch on caudal-fin base; lateral line reaching a vertical through
	adipose-fin origin
_	Triangle-shaped blotch on caudal-fin base; lateral line reaching middle of
	dorsal-fin base

### Pimelodidae

80

1	Upper jaw projected well beyond lower jaw with premaxillary tooth patch exposed <b>Sorubim lima</b>
_	Lower jaw just slightly or not projected at all, premaxillary tooth plate not
	completely exposed
2	Upper portion of caudal-fin base with well-defined dark blotch
_	Caudal-fin base without well-defined dark blotch
3	Leading pectoral-fin ray soft and not pungent or spinous
	Megalonema platycephalum
-	Leading pectoral-fin ray strong, pungent, with anterior and posterior serrae.
,	
4	Eye visible in ventral view, set below midline of head in lateral view; barbels
	flat and wide
-	Eye not visible in ventral view, set above midline in lateral view; barbels round
_	in cross-section
5	Body color pattern consisting of dark vertical bars
_	Body color pattern consisting of dark spots or plain coloration, never with vertical bars7
6	Abundant (more than 50) spots on caudal-fin Pseudoplatvstoma metaense
	Few (less than 50) spots on caudal fin <b>Devidablatistama aminacoansa</b>
_	Tew (less than )0) spots on cautal-infi
7	Body color pattern consisting of numerous spots or blotches, sometimes fuss-
7	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
7 	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
7	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 -	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 -	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9 -	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9 - 10	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9 - 10	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9 - 10	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat
- 7 - 8 - 9 - 10 -	Body color pattern consisting of numerous spots or blotches, sometimes fuss- ing so that a vermicular pattern appears; interorbital region flat

## Auchenipteridae

1	Mental barbels absent	2
_	Mental barbels present	3
2	Caudal fin deeply forkedAge	eneoisus ucayalensis
_	Caudal fin truncate	Ageneiosus magoi
3	7–9 anal-fin raysCen	tromochlus romani
_	>18 anal-fin rays	4
4	Dorsal-fin spine shorter than pectoral-fin spine Pseude	epapterus hasemani
_	Dorsal-fin spine longer than or equal to pectoral-fin spir	ne5
5	Caudal fin obliquely truncated Track	belyopterus galeatus
_	Caudal fin forked	ntomocorus gameroi

### **CHARACIFORMES**

1	Mouth without teeth Curimatidae
_	Mouth with teeth2
2	Canine teeth present
_	Canine teeth absent6
3	Adipose fin absent; caudal fin roundedErythrinidae
_	Adipose fin present; caudal fin not rounded4
4	Dentary canine teeth hypertrophied; caudal fin truncated; rostrum short
	Ctenoluciidae
-	Dentary without hypertrophied canine teeth; caudal fin bifurcated; rostrum
	large
5	Scales ctenoid Ctenolucidae
_	Scales cicloidAcestrorhynchidae
6	Upper jaw without teeth; upper lip with teeth7
_	Upper jaw with teeth; upper lip without teeth
7	Predorsal process present; mouth evertible
	Prochilodontidae: Prochilodus mariae
_	Predorsal process absent; mouth non-evertible Chilodontidae
8	Abdominal serrated keel present Serrasalmidae
_	Abdominal serrated keel absent9
9	Branchial membrane fused to isthmus Anostomidae
-	Branchial membrane free from isthmus10
10	Pectoral fins strongly developed and horizontally oriented11
_	Pectoral fins vertically oriented12
11	Fewer than three unbranched rays in pectoral fin Parodontidae
_	Three unbranched rays in pectoral fin Crenuchidae
12	Chest extremely compressed and expanded, forming a ventral keel
_	Chest not expanded to form ventral keel14

13	Anal-fin origin anterior to dorsal-fin origin; ventral keel well developed
	Gasteropelecidae: Thoracocharax stellatus
_	Anal-fin origin posterior to dorsal-fin origin; ventral keel moderately devel-
	opedTriportheidae
14	Upper lobe of caudal fin longer than lower lobe Lebiasinidae
_	Both lobes of caudal fin of equal size15
15	Two rows of teeth in the dentary Bryconidae
_	One row of teeth in dentary16
16	Anterior margin of maxilla greatly arched above and meeting premaxilla bor-
	der at right angle Iguanodectidae: Bryconops giacopinni
_	Anterior margin of maxilla not greatly arched above and not meeting premax-
	illa border at right angleCharacidae

### Curimatidae

1	Lateral line incomplete, with only few pored scales
_	Lateral line complete with all or almost all lateral scales with pores
2	Caudal fin with scales reaching two thirds in length in both lobes
_	Caudal fin with scales only on the base of caudal-fin rays
3	Lateral line with 42 or more scales
_	Lateral line with 39 or fewer scales
4	Lateral-line scales more than 90 Potamorhina altamazonica
_	Lateral-line scales 42–60
5	Mouth terminal; dorsal fin without dark spot
_	Mouth inferior; dorsal fin with or without dark spot7
6	Interorbital width 53-58% of HL; anal-fin rays iii,10-iii,12.; abdominal re-
	gion, pectoral, pelvic and anal fins with red coloration in live specimens
	Curimata cerasina
_	Interorbital width less than 52% of HL. Anal-fin rays iii,8-iii,9; abdominal
	region, pectoral, pelvic and anal fins without red coloration in live speci-
	mensPsectrogaster ciliata
7	Dark spot on the dorsal fin; prepelvic region of body not flattened or only
	obtusely flattened; 1 to 3 scales between posterior border of anus and anal-fin
	originSteindachnerina pupula
-	No dark spot on the dorsal fin; prepelvic region of body distinctly flattened;
	5 to 7 scales between posterior border of anus and anal-fin origin
	Steindachnerina hypostoma
8	Without a dark spot on the basal portion of the middle rays of the dorsal fin
	Cyphocharax spilurus
_	With a dark spot on the basal portion of the middle rays of the dorsal fin
	Steindachnerina argentea

#### Erythrinidae

1	Maxilla with 2-3 canine teeth; caudal fin usually spot	ted; dark lateral stripe
	absent or diffuse	Hoplias malabaricus
_	Maxilla without canine teeth; caudal fin not spotted;	dark lateral stripe very
	well marked Hoplery	ythrinus unitaeniatus

### Cynodontidae

1	Dorsal-fin origin anterior to	anal-fin origin	Hydrolycus armatus
_	Dorsal-fin origin in line with	h anal-fin origin	Raphiodon vulpinus

### Ctenoluciidae

1	In adults, all lateral-line scales perforated (82 or more); body without round
	spots, except one at the base of caudal finBoulengerella cuvieri
_	In adults, only up to 25 lateral-line scales perforated; body with black spots.

### Acestrorhynchidae

1	Lateral line complete with 99–13	l scales; dorsal-fin origin anterior to the
	anal-fin origin	Acestrorbynchus microlepis
_	Lateral line incomplete with 33-37	lateral-line scales; dorsal-fin origin poste-
	rior to the anal-fin origin	Gnathocharax steindachneri

### Chilodontidae

## Serrasalmidae

1	Adipose-fin base long, its length longer than the distance between posterior
	margin of dorsal-fin base to the adipose-fin originMetynnis argenteus
-	Adipose-fin base short, its length shorter than the distance between posterior
	margin of dorsal-fin base to the adipose-fin origin2
2	Two rows of teeth on the premaxilla Mylossoma duriventre
_	One row of teeth on the premaxilla
3	Base of dentary teeth circular and separated from adjacent ones; lower jaw prom-
	inent and projecting greatly forward from upper jaw Catoprion mento
_	Base of dentary teeth flattened, in contact or very close to adjacent ones;
	lower jaw equal or slightly anterior to upper jaw
4	29 or fewer anal-fin rays; snout, head, jaws, and body short and robust; dorsal
	profile of head to posterior margin of eyes convex; prepelvic and abdominal
	region red in life; black and conspicuous humeral blotch posterior to oper-
	clePygocentrus cariba
_	30 or more anal-fin rays; snout, head, jaws, and body long and slender; dorsal
	profile of head to posterior margin of eyes concave; prepelvic and abdominal
	region red only during breeding season
5	Base of posterior premaxillary tooth (6th) approximately equal to adjacent
	one (5th); ectopterygoid teeth unicuspid, generally 5 or fewer
	Pristobrycon striolatus
_	Base of posterior premaxillary tooth (6th) wider than adjacent one (5 <sup>th</sup> ); ec-
	topterygoid teeth bicuspid or tricuspid, generally 7 or more
6	Posterior margin of caudal fin hyaline, with black vertical bar at base of cau-
	dal finSerrasalmus irritans
_	Caudal fin completely black or with its posterior margin black7
7	Body width 1.7 or less in SL; lateral spots vertically elongated
	Serrasalmus altuvei
_	Body width 1.7–2.0 in SL; lateral spots rounded Serrasalmus rhombeus
	• •

### Anostomidae

1	Caudal fin with scales only on its base and without dark bars2
_	Caudal fin with scales at least over two-thirds of both lobes. Caudal-fin with
	oblique dark bars
2	Dentary teeth tricuspid to pentacuspid; 7 branched anal-fin rays
	Schizodon scotorhabdotus
_	Dentary teeth incisiform or bicuspid; 8 or more branched anal-fin rays3
3	Body with seven dark vertical bars, the second with a "Y" shape
	Leporinus y-ophorus
_	Body with rounded spots or longitudinal stripes

Body with 4–5 longitudinal stripes separated by white or yellow stripes......
 *Leporinus striatus* Body with two rounded black spots at midline ....... *Leporinus* gr. *friderici*

### Parodontidae

Dark vertical bars present; dark lateral stripe absent ....... Parodon apolinari
 Dark vertical bars absent; dark lateral stripe present..... Parodon aff. suborbitalis

### Crenuchidae

1	Chest naked without scales between pectoral fins Characidium gr. boavistae
_	Chest covered with scales between pectoral fins2
2	Body pigmentation composed by irregular lines and dots Characidium pteroides
_	Body pigmentation composed by well-defined vertical bars
3	9 or fewer branched pectoral-fin rays
_	10 or more branched pectoral-fin rays5
4	All vertical bars always originating on dorsum; body depth 23% of HL or
	less
_	Not all vertical bars originating on dorsum, thin vertical bars not reaching the
	dorsum between vertical bars originating on dorsum; body depth 24% of HL
	or more
5	Mid-lateral stripe diffuse or absent; vertical bars fragmented in rhomboidal
	shape Characidium cf. steindachneri
_	Mid-lateral stripe very well developed; vertical bars continuous and with dots
	appearance at junction with mid-lateral line Characidium chupa

### Triportheidae

1	Pectoral fin not surpassing ventral-fin originEngraulisoma taeniatum
_	Pectoral fin surpassing ventral-fin origin2
2	24–27 branched anal-fin rays; 5–6 scale rows between lateral line and dorsal-
	fin origin <i>Triportheus venezuelensis</i>
2	28–32 branched anal-fin rays; 7 scale rows between lateral line and dorsal-fin
	origin

### Lebiasinidae

1	Adipose fin present	Lebiasina erythrinoid	es
_	Adipose fin absent		.2

### Bryconidae

1	Two rows of teeth in the premaxilla	Salminus sp.
_	Three rows of teeth in the premaxilla	2
2	Dark lateral stripe present, from opercle to tip of middle ca	audal-fin rays; up-
	per caudal-fin lobe unpigmented	Brycon whitei
_	Dark lateral stripe absent; upper caudal-fin lobe pigmented.	Brycon falcatus

### Characidae

1	Lateral line incomplete
_	Lateral line complete
2	Dorsal fin with a black blotch
_	Dorsal fin without blotches
3	Body height 26.6 to 29.3% of SL Hyphessobrycon dorsalis
_	Body height is 34.5 to 36.9% of SL Hyphessobrycon sweglesi
4	One row of premaxillary teeth
_	Two rows of premaxillary teeth7
5	Dorsal-fin origin posterior to the anal-fin origin Paragoniates alburnus
_	Dorsal-fin origin anterior to the anal-fin origin
6	A dark embedded crescent-shaped mark on base of each caudal lobe; no red
	color on caudal fin Microschemobrycon casiquiare
_	Without such dark pigmentation on the base of caudal fin; caudal fin red in
	life Aphyocharax alburnus
7	Caudal fin with black pigmentation in both lobes
	Hyphessobrycon otrynus
-	Caudal fin without black pigmentation
8	
	Anal-fin base with a conspicuous dark band Hemigrammus barrigonae
_	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation9
- 9	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation
- 9	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation
_ 9 _	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation
- 9 - 10	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation
- 9 - 10	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation
9 10 	Anal-fin base with a conspicuous dark band <i>Hemigrammus barrigonae</i> Anal-fin base without dark pigmentation

11	With a conspicuous caudal spot12
_	Without a caudal spot or with a lateral stripe continuing along middle cau-
	dal-fin rays, but not forming a caudal spot14
12	Dark melanophores present on sides of body and between anal fin and lateral
	No dark melanophores present on sides of body and between anal fin and
_	lateral line
13	Caudal spot triangle-shaped covering the central portion of caudal-fin base,
	continuing along middle caudal-fin rays and usually not in contact with the
	lateral stripe
_	A large caudal spot covering entire caudal-fin base, continuing along middle
1 /	caudal-fin rays and in contact with lateral stripe <i>Hemigrammus newboldi</i>
14	Body depth 2/% or less of SL; 14–16 branched anal-fin rays; maxillary teeth
	absent; black blotch on upper caudal-fin lobe <i>Hyphessobrycon diancistrus</i>
_	Body depth 28% or more of SL; 18–20 branched anal-fin rays; maxillary
	teeth present; without dark blotch on upper caudal-fin lobe
15	12 dentary teeth; 6–7 perforated lateral-line scales. Without a dark blotch at
	caudal peduncle
_	14 dentary teeth; 9–10 perforated lateral-line scales. With a dark blotch at
	caudal peduncle
16	Dorsal-fin origin at or posterior to vertical through anal-fin origin17
-	Dorsal-fin origin anterior to the anal-fin origin
17	61–68 anal-fin rays
_	25–55 anal-fin rays
18	Length of maxilla equal or shorter than vertical diameter of eye; adult males
	with hypertrophied caudal-fin squamation on lower caudal-fin lobe
	Gephyrocharax valencia
-	Length of maxilla longer than vertical diameter of eye; adult males without
	hypertrophied caudal-fin squamation19
19	No external mammiliform teeth Charax sp.
_	External mammiliform teeth present in the maxilla and premaxilla
20	50–65 lateral-line scales
_	66 or more lateral-line scales
21	66–70 perforated lateral-line scales; 12–14 gill rakers on lower arm of the first
	gill arch
21	70-88 perforated lateral-line scales; 7-11 gill rakers on lower arm of the first
	gill arch
22	16 or less total anal-fin rays23
_	17 or more total anal-fin rays27
23	Premaxillary teeth arranged in 2 rows; Anterior triad of larger teeth with
	rounded base absent
_	Premaxillary teeth arranged in 3 rows; Anterior triad of larger teeth with
	rounded base present

<ul> <li>Dorsal-fin origin posterior or aligned with pelvic-fin origin</li></ul>	24	Dorsal-fin origin anterior to pelvic-fin insertion Creagrutus sp.
<ul> <li>Third infraorbital poorly developed, leaving a broad gap between its posterior margin and the horizontal limb of preopercle</li></ul>	-	Dorsal-fin origin posterior or aligned with pelvic-fin origin25
<ul> <li>margin and the horizontal limb of preopercle</li></ul>	25	Third infraorbital poorly developed, leaving a broad gap between its posterior
-       Ihird infraorbital well-developed contacting or with a small gap from horizontal limb of preopercle       26         26       38-42 lateral-line scales; interorbital distance 34.0-37.9% of HL       26         27       36-38 lateral-line scales; interorbital distance 28.6-35.4% of HL       28         28       39 or less total anal-fin rays       28         29       Creagrutus cf. taphorni         20       Xeloid scales on body sides.       29         29       Ctenoid scales on body sides, or at least in preventral area       30         29       Maxilla shorter than vertical diameter of eye; cleithrum without a notch on posteroventral portion; body sides with dark sinuous linesMarkiana geayi         -       Maxilla longer than vertical diameter of eye; cleithrum without a notch on posteroventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines         30       S8 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid; ctenoid scales only on preventral area       Charax metae         31       100-110 lateral-line scales       32         32       35-38 branched anal-fin rays; 73-76 lateral-line scales       32         33       39 or cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle       Exodon paradoxus         34       One row of		margin and the horizontal limb of preopercle Creagrutus bolivari
zontal limb of preopercle       26         38-42 lateral-line scales; interorbital distance 34.0-37.9% of HL	-	Third infraorbital well-developed contacting or with a small gap from hori-
26       38–42 lateral-line scales; interorbital distance 34.0–37.9% of HL.	26	zontal limb of preopercle
<ul> <li><i>Creagrutus atratus</i></li> <li>36–38 lateral-line scales; interorbital distance 28.6–35.4% of HL</li> <li><i>Creagrutus</i> cf. <i>taphorni</i></li> <li>40 or more total anal-fin rays.</li> <li>39 or less total anal-fin rays.</li> <li>28 Gycloid scales on body sides.</li> <li>29 Ctenoid scales on body sides, or at least in preventral area</li> <li>30 Maxilla shorter than vertical diameter of eye; cleithrum without a notch on posteroventral portion; body sides with dark sinuous lines<i>Markiana geayi</i></li> <li>Maxilla longer than vertical diameter of eye; cleithrum with notch on posteroventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines<i>Charax metae</i></li> <li>58 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid; ctenoid scales only on preventral area<i>Ctenobrycon spilurus</i></li> <li>59 or more lateral-line scales; one or more conical or canine teeth; ctenoid scales on entire body</li></ul>	26	38–42 lateral-line scales; interorbital distance 34.0–3/.9% of HL
27       40 or more total anal-fin rays       28         29       39 or less total anal-fin rays       33         28       Cycloid scales on body sides, or at least in preventral area       30         29       Ctenoid scales on body sides, or at least in preventral area       30         29       Maxilla shorter than vertical diameter of eye; cleithrum without a notch on posteroventral portion; body sides with dark sinuous lines <i>Markiana geayi</i> -       Maxilla longer than vertical diameter of eye; cleithrum with notch on posteroventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines <i>Charax metae</i> 30       58 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid; ctenoid scales only on preventral area <i>Ctenobrycon spilurus</i> -       59 or more lateral-line scales; one or more conical or canine teeth; ctenoid scales on entire body       31         31       100–110 lateral-line scales <i>Cynopotamus bipunctatus</i> -       70–84 lateral-line scales       32         32       35–38 branched anal-fin rays; 73–76 lateral-lines scales <i>Galeocharax gulo</i> 33       A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle <i>Exodon paradoxus</i> 34       One row of premaxillary teeth	_	36–38 lateral-line scales; interorbital distance 28.6–35.4% of HL
27       40 or more total anal-fin rays       28         39 or less total anal-fin rays       33         28       Cycloid scales on body sides.       29         -       Ctenoid scales on body sides, or at least in preventral area       30         29       Maxilla shorter than vertical diameter of eye; cleithrum without a notch on posteroventral portion; body sides with dark sinuous lines       30         29       Maxilla longer than vertical diameter of eye; cleithrum with notch on posteroventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines <i>Charax metae</i> 30       58 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid; ctenoid scales only on preventral area. <i>Ctenobrycon spilurus</i> -       59 or more lateral-line scales; one or more conical or canine teeth; ctenoid scales on entire body.       31         31       100–110 lateral-line scales <i>Cynopotamus bipunctatus</i> -       70–84 lateral-line scales       32         32       35–38 branched anal-fin rays; 73–76 lateral-line scales <i>Galeocharax gulo</i> 33       A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle <i>Exodon paradoxus</i> -       No cuspidate teeth on premaxilla; different color pattern than that described above       3		
<ul> <li>39 or less total anal-fin rays</li></ul>	27	40 or more total anal-fin rays
<ul> <li>Cycloid scales on body sides</li></ul>	_	39 or less total anal-fin rays
<ul> <li>Ctenoid scales on body sides, or at least in preventral area</li></ul>	28	Cycloid scales on body sides
<ul> <li>Maxilla shorter than vertical diameter of eye; cleithrum without a notch on posteroventral portion; body sides with dark sinuous lines Markiana geayi</li> <li>Maxilla longer than vertical diameter of eye; cleithrum with notch on posteroventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines</li></ul>	-	Ctenoid scales on body sides, or at least in preventral area
<ul> <li>posteroventral portion; body sides with dark sinuous lines Markiana geayi</li> <li>Maxilla longer than vertical diameter of eye; cleithrum with notch on poster- oventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines</li></ul>	29	Maxilla shorter than vertical diameter of eye; cleithrum without a notch on
<ul> <li>Maxilla longer than vertical diameter of eye; cleithrum with notch on poster- oventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines</li></ul>		posteroventral portion; body sides with dark sinuous lines Markiana geayi
oventral portion, near base of posteriorly directed spiniform projection; body sides with dark sinuous lines	-	Maxilla longer than vertical diameter of eye; cleithrum with notch on poster-
<ul> <li>sides with dark sinuous lines</li></ul>		oventral portion, near base of posteriorly directed spiniform projection; body
30       58 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid; ctenoid scales only on preventral area		sides with dark sinuous lines Charax metae
<ul> <li>ctenoid scales only on preventral area</li></ul>	30	58 or fewer lateral-line scales; premaxillary and dentary teeth multicuspid;
<ul> <li>59 or more lateral-line scales; one or more conical or canine teeth; ctenoid scales on entire body</li></ul>		ctenoid scales only on preventral areaCtenobrycon spilurus
scales on entire body       31         31       100–110 lateral-line scales <i>Cynopotamus bipunctatus</i> -       70–84 lateral-line scales       32         32       35–38 branched anal-fin rays; 73–76 lateral-line scales       32         33       A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle <i>Exodon paradoxus</i> -       No cuspidate teeth on premaxilla; different color pattern than that described above       34         0ne row of premaxillary teeth       35         -       Two rows of premaxillary teeth       38	-	59 or more lateral-line scales; one or more conical or canine teeth; ctenoid
<ul> <li>31 100–110 lateral-line scales</li></ul>		scales on entire body
<ul> <li>70–84 lateral-line scales</li></ul>	31	100–110 lateral-line scales <i>Cynopotamus bipunctatus</i>
<ul> <li>32 35–38 branched anal-fin rays; 73–76 lateral-line scales</li></ul>	-	70–84 lateral-line scales
<ul> <li>Acestrocephalus sardina</li> <li>39–45 branched anal-fin rays; 79–84 lateral-lines scales Galeocharax gulo</li> <li>A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle</li></ul>	32	35–38 branched anal-fin rays; 73–76 lateral-line scales
<ul> <li>39–45 branched anal-fin rays; 79–84 lateral-lines scales <i>Galeocharax gulo</i></li> <li>A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle</li></ul>		Acestrocephalus sardina
<ul> <li>A pair of cuspidate teeth on premaxilla pointing forward on labial sides of upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle</li></ul>	_	39–45 branched anal-fin rays; 79–84 lateral-lines scales Galeocharax gulo
<ul> <li>upper jaw; two large black blotches on body sides, one anterior to dorsal-fin origin and another on caudal peduncle</li></ul>	33	A pair of cuspidate teeth on premaxilla pointing forward on labial sides of
<ul> <li>origin and another on caudal peduncle</li></ul>		upper jaw; two large black blotches on body sides, one anterior to dorsal-fin
<ul> <li>No cuspidate teeth on premaxilla; different color pattern than that described above</li></ul>		origin and another on caudal peduncle <i>Exodon paradoxus</i>
<ul> <li>34 One row of premaxillary teeth</li></ul>	_	No cuspidate teeth on premaxilla; different color pattern than that described above
- Two rows of premaxillary teeth	34	One row of premaxillary teeth
	_	Two rows of premaxillary teeth
55 Premaxillary teeth with five cusps, the outer cusps very small; dentary teeth	35	Premaxillary teeth with five cusps, the outer cusps very small; dentary teeth
with five cusps, the three central cusps flat and approximately equal in size		with five cusps, the three central cusps flat and approximately equal in size
and the outer cusps very small Cheirodontops geayi		and the outer cusps very small Cheirodontops geayi
- Premaxillary teeth with nine cusps; dentary teeth heptacuspid, with central	_	Premaxillary teeth with nine cusps; dentary teeth heptacuspid, with central
cusp longer than others		cusp longer than others
36 A remarkable elongation of the second unbranched dorsal-fin ray in males; maxilla somewhat triangular short with mid-length portion deeper and	36	A remarkable elongation of the second unbranched dorsal-fin ray in males; maxilla somewhat triangular short, with mid-length portion deeper and
gradually narrowing to the posterior tip		gradually narrowing to the posterior tip

_	No elongation of the second unbranched dorsal-fin ray in males; maxilla somewhat spatula-like shaped, short or elongate, with a deep mid-length re-
37	Adipose-fin origin at vertical through second or third last anal-fin ray inser-
	rays
_	Adipose-fin origin at vertical through last anal-fin ray insertion: mature males
	with hooks on first to twenty second branched anal-fin rays
	Odontostilbe pulchra
38	Presence of a predorsal spine; anterior three anal-fin rays black
	Poptella compressa
_	No predorsal spine; three anterior anal-fin rays unpigmented
39	33–38 total anal-fin rays
_	32 or less total anal-fin rays
40	Body depth 28.0-37.0% of SL; maxilla with 20-30 conical teeth; 6 scale
	rows between lateral line and dorsal-fin origin; one humeral spot
	Phenacogaster maculoblonga
_	Body depth 38.0% or more of SL; maxilla with 1–3 multicuspidate teeth; 7–9
	scale rows between lateral line and dorsal-fin origin; two humeral spots
41	Caudal peduncle with a large dark spot
_	Caudal peduncle without spot Gymnocorymbus bondi
42	16–20 total anal-fin rays
_	23–32 total anal-fin rays
43	17–18 anal-fin rays Knodus deuterodonoides
_	16 anal-fin raysBryconamericus cf. cismontanus
44	Spine-like pelvic bones projecting anteriorly from pelvic-fin base, with distal
	tip free from musculature <i>Jupiaba polylepis</i>
-	No spine-like pelvic bones
45	4 teeth on the inner row of premaxilla
-	5 teeth on the inner row of premaxilla
46	3 or fewer teeth on maxilla
_	More than three teeth on maxilla
47	5 or fewer maxillary teeth in adult individuals (>5 cm SL); second premaxil-
	lary tooth of inner row pentacuspid; 12–13 predorsal scales; fins of live speci-
	mens reddish
-	6 or more maxillary teeth in adult individuals (>5 cm SL); second premaxil-
	lary tooth of inner row heptacuspid; 14–16 predorsal scales; fins of live speci-
<i>(</i> -	mens not reddish Hemibrycon metae
48	One or both caudal-fin lobes with dark coloration
-	Both caudal-fin lobes without distinct pigmentation
49	Only upper caudal-fin lobe with dark pigmentation
	Moenkhausia lepidura
-	Both caudal-fin lobes with black pigmentation Moenkhausia dichoura

50	Middle caudal-fin rays without dark coloration	Moenkahusia copei
_	Middle caudal-fin rays with dark coloration	
51	Predorsal line naked	Astyanax bimaculatus
_	Predorsal line with scales	
52	Base of anal fin with an oblique dark stripe extendin	ng across caudal peduncle
	and onto middle and upper caudal-fin rays	Astyanax metae
_	Base of anal fin without oblique dark stripe	
53	45 or more lateral-line scales	Astyanax integer
_	Fewer than 45 lateral-line-scales	Astyanax venezuelae

## Cichlidae

1	African lips (posterior portion of the lower lip not covering part of the up per lip): three roundish ocellar blotches in adults (large and oval blotches it	
	juveniles)	
_	American lips (posterior portion of the lower lip covering part of the upper lip); different color pattern than above	
2	Bone expansion in the upper region of the first gill arch forming a well- developed fleshy lobe	
_	First gill arch without such a lobe6	
3	Lower pharyngeal bone without teeth along its margin	
_	Lower pharyngeal bone with teeth along its margin	
4	Lateral spots on body present; two pectoral spots <i>Apistogramma macmasteri</i>	
_	No lateral spots on body; with or without a single pectoral spot	
5	Pectoral fin base with spot; with distinct abdominal stripes	
	Apistogramma hongsloi	
-	Without spot on pectoral fin base; without abdominal stripes	
	Apistogramma hoignei	
6	Irregular predorsal scale pattern7	
_	Uniserial or triserial predorsal scale pattern	
7	Six or seven anal-fin spines; 24–30 scales in upper lateral line series; vertical	
	bars 6 and 7 parallel; body deep (50.7–55.6% of SL) Mesonauta egregius	
-	Three anal-fin spines; 40 or more scales in upper lateral line series; elongate	
0	and somewhat cylindrical body (less than 50.0% of 5L)8	
8	Humeral blotch present	
-	Humeral blotch absent	
9	Lateral line crossing under portion of humaral blotch	
10	Chain of blotches along lateral lines: length of posterior dorsal fin spine	
10	9 6–10 6% of SL: caudal peduncle length at ventral part 10 0–11 5% of SL:	
	length of ventral fin 19.4–20.4% of SL <i>Crenicichla sveni</i>	
	0	

_	No chain of blotches along lateral lines; length of posterior dorsal-fin spine
	10.8–11.2% of SL; caudal peduncle length at ventral part 11.8–12.2% of SL;
	length of ventral fin 17.3–18.2% of SL Crenicichla sp.
11	Maxilla extending posterior to the anterior margin of eye; caudal-fin length
	(from caudal-fin base to tips of middle caudal-fin rays) 16.2-24.6% of SL;
	head depth at orbit level 16.7-21.6% SL; caudal peduncle depth where least
	10.7–14.2% of SL <i>Crenicichla geayi</i>
_	Maxilla only reaching anterior margin of eye; caudal-fin length (from caudal-
	fin base to tips of middle caudal-fin rays) 22.7–24.7% of SL; head depth at
	orbit level 15.1–15.6% SL; caudal peduncle depth where least 9.34–9.83%
	of SL Crenicichla gr. wallacii
12	Uniserial predorsal scale pattern
_	Triserial predorsal scale pattern14
13	8 branched anal-fin rays; caudal fin rounded; less than 8 scales in the lower
	lateral line Andinoacara sp.
-	7 branched anal-fin rays; caudal fin subtruncate-truncate; more than 8 scales
	in the lower lateral lineBujurquina mariae
14	Dark stripe extending from posterodorsal margin of eye to lower angle of
	preopercle
-	Dark stripe restricted to a suborbital blotch only14
15	Head sides with bluish or greenish iridescent stripes (in live specimens)
	Aequidens diadema
-	Head sides without bluish or greenish iridescent stripes (in live specimens)
	Aequidens tetramerus

### Sciaenidae

### Discussion

Regional checklists of freshwaters fishes become dynamic over time as studies in freshwater fish taxonomy and distribution advance for the Neotropical region (Reis et al. 2016). The most recent checklist for the Meta River basin reported 258 species for the Cusiana River sub-basin (Usma-Oviedo et al. 2016); it is a higher number of species than reported herein because re-identification and taxonomic updating processes of specimens excluded 59 of the 258 nominal species reported by Usma-Oviedo et al. (2016). For example, most of the undetermined (e.g., *Ancistrus* sp., *Aphyocharax* sp, *Microglanis* sp., and *Odontostilbe* sp.) and erroneous records (e.g., *Hemibrycon cristiani*, *Pyrrhulina brevis*, *Schultzichthys gracilis*, and *Steindachnerina guentheri*) originally counted as independent species were merged with other recorded species after our verification of the data. This is not surprising, taxonomy proceeds at a faster pace than the institutional ability to maintain updated records. For this reason, there is a need to account for validation of species identification when regional checklists are assembled from multiple secondary sources in order to avoid errors due to outdated or unverified data.

Extrapolation suggests that the drainage could have a richness of roughly 314 species, indicating that the number of species found in the present study represents 77.7% of the expected richness in the area (Table 1). However, this estimate represents a rough estimate because sampling efforts have not been uniform across the drainage. A historical sampling-specific bias is not expected in the Cusiana River sub-basin, with the possible exception of an elevational bias (as is true for the whole Orinoco drainage in Colombia). Given the non-uniform nature of sampling in comparable river systems, we suggest that our extrapolations of species richness may be useful for comparisons among drainages using collection records as the input for rough estimation. This is particularly important since most checklist studies compare observed and not estimated richness, with the latter a more appropriate measure because it incorporates (and even overestimates) uncertainty from the samples into the estimation, and it also serves accounts for sampling effort among drainages.

Among similar Orinoco Andean tributaries, the Cusiana is one of the best-sampled sub-basins, exceeding in species richness other recently well-sampled sub-basins such as Orotoy (113 spp.; Ramírez-Gil et al. 2011) and Pauto (182 spp.; Maldonado-Ocampo et al. 2013). In fact, the Cusiana River sub-basin represents the quantile 0.83 among Orinocoan drainages, indicating that 83 % of the other drainages had fewer than 74 localities represented in the collections. The importance of the Cusiana River sub-basin is not only determined by its fish richness, but also because of its diverse and extensive aquatic ecosystem richness (rivers, streams, lagoons, estuaries, palm swamps, riparian forests, and flooded savannas) that provide important areas for fish reproduction, shelter and food. Because aquatic ecosystems have dynamic ecological and environmental processes (Teresa et al. 2015, Ribeiro et al. 2016, Toussaint et al. 2016), management and conservation projects of sub-basins should be addressed at regional (sub-basin) scales.

The documentation of the ichthyofauna in cis-Andean Colombian sub-basins has been increasing during the last decade, but new records and species can likely still be found in areas previously thought to be well-sampled (e.g., Ballen 2011, Vanegas-Ríos et al. 2015, Ballen et al. 2016a, 2016b, Burns et al. 2017, García-Alzate et al. 2017). Most of the sampling effort has been carried out in the piedmont and lowland areas in the Cusiana as well as in other sub-basins, and exploration of High Andean areas could lead to the discovery of local endemic species at the basin scale that usually are underestimated (Carvajal-Quintero et al. 2015).

Sub-basins adjacent to the Cusiana draining along the eastern slope of the Eastern Cordillera in the Orinoco region of Colombia (e.g., Guachiría, Casanare, Upía, Túa, and Cravo Sur) have not been well sampled and their richness is surely underestimated (Urbano-Bonilla et al. 2014). Continuous efforts are still to be carried out in order to document the fish fauna present along this region; this information is crucial to better understand how different anthropogenic activities (mining, oil extraction, agricultural, and livestock practices) are affecting the environmental conditions of these areas and as a consequence, the fish that live therein. Combination of this kind of information and further environmental data is a necessary step in order to generate freshwater conservation strategies using different approaches and therefore go further toward effective protection initiatives for species subject of conservation in the region.

#### Acknowledgments

Funding to conduct field work along the Orinoco piedmont were provided by the Pontificia Universidad Javeriana (Project N° 5211411), Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, and Oleoducto Bicentenario (Project N° 4400000166); cooperation agreement between Fundación Reserva Natural La Palmita, Centro de Investigación and IAvH (N° 15-14-172-010CE), and Gobernación de Casanare (Consulting contract N° 1819-2014). GAB was funded through a doctoral scholarship and a BEPE internship by FAPESP (processes 2014/11558-5 and 2016/02253-1). To Dr. Francisco Villa-Navarro for providing fish database of the Zoological Collection of the University of Tolima (CZUT-IC). The authors thank Carlos A. García-Alzate, Donald Taphorn, Flavio Lima, Henry Agudelo, Jorge E. García-Melo, Paulo Andreas Buckup, and Vitor Abrahão for their valuable time and comments on taxonomic verification of species or keys, and María Paula Arango and Natalia Borray for their valuable support in some phases of the laboratory analysis. We acknowledge Donald Taphorn and Devin Bloom for checking the English of the manuscript.

#### References

Abell R, Thieme ML, Revenga C, Bryer M, Kottelat M, Bogutskaya N, Coad B, Mandrak M, Contreras Balderas S, Bussing M, Stiassny MLL, Skelton P, Allen GR, Unmack P, Naseka A, Sindorf R, Ng N, Robertson J, Armijo E, Higgins JV, Heibel TJ, Wikramanayake E, Olson D, López HL, Reis RE, Lundberg JV, Sabaj-Pérez MH, Petry P (2008) Freshwater ecoregions of the World: a new map of biogeographic units for freshwater biodiversity conservation. BioScience 58: 403–414. https://doi.org/10.1641/B580507

- Anderson EP, Maldonado-Ocampo JA (2011) A regional perspective on the diversity and conservation of tropical Andean fishes. Conservation Biology 25: 30–39. https://doi:10.1111/ j.1523-1739.2010.01568.x
- Ballen GA (2011) A new species of *Chaetostoma* Tschudi (Siluriformes: Loricariidae) from Colombia with a definition of the *C. anale* species group. Papéis Avulsos de Zoologia (São Paulo) 51(26): 383–398. http://dx.doi.org/10.1590/S0031-10492011002600001
- Ballen GA, Urbano-Bonilla A, Maldonado-Ocampo JA (2016a) Description of a new species of the genus *Chaetostoma* from the Orinoco River drainage with comments on *Chaetostoma milesi* Fowler, 1941 (Siluriformes: Loricariidae). Zootaxa 4105: 181–197. http://dx.doi. org/10.11646/zootaxa.4105.2.6
- Ballen GA, Urbano-Bonilla A, Zamudio JE (2016b) *Farlowella mitoupibo*, a new species of stick catfish from the upper Guaviare River, Orinoco basin, Colombia (Teleostei: Loricariidae). Ichthyological Exploration of Freshwaters 27: 325–332.
- Barletta M, Jaureguizar AJ, Baigun C, Fontoura NF, Agostinho AA, Almeida-Val VMF, Val AL, Torres RA, Jiménez-Segura LF, Giarrizzo T, Fabré NN, Batista VS, Lasso C, Taphorn D, Costa MF, Chaves PT, Vieira JP, Correa MFM (2010) Fish and aquatic habitat conservation in South America: a continental overview with emphasis on Neotropical systems. Journal of Fish Biology 76: 2118–2176. https://doi.org/10.1111/j.1095-8649.2010.02684.x
- Betancur-R R, Wiley E, Bailly N, Miya M, Lecointre G, Ortí G (2016) Phylogenetic Classification of Bony Fishes. https://sites.google.com/site/guilleorti/classification-v-4 [accessed 31 Jul 2017]
- Burns MD, Chatfield M, Birindelli JL, Sidlauskas BL (2017) Systematic assessment of the *Leporinus desmotes* species complex, with a description of two new species. Neotropical Ichthyology 15(2). http://dx.doi.org/10.1590/1982-0224-20160166
- Carvajal-Quintero JD, Escobar F, Alvarado F, Villa-Navarro FA, Jaramillo-Villa Ú, Maldonado-Ocampo JA (2015) Variation in freshwater fish assemblages along a regional elevation gradient in the northern Andes, Colombia. Ecology and Evolution 5: 2608–2620. http:// doi:10.1002/ece3.1539
- Chao A, Gotelli NJ, Hsieh TC, Sander EL, Ma KH, Colwell RK Ellison AM (2014) Rarefaction and extrapolation with Hill numbers: a framework for sampling and estimation in species diversity studies. Ecological Monographs 84: 45–67. https://doi.org/10.1890/13-0133.1
- DoNascimiento C, Herrera-Collazos EE, Herrera-R GA, Ortega-Lara A, Usma-Oviedo JS, Villa-Navarro F, Maldonado-Ocampo JA (2017) Checklist of the freshwater fishes of Colombia: a Darwin Core alternative to the updating problem. ZooKeys 708: 25–138. https:// doi.org/10.3897/zookeys.708.13897
- Eschmeyer WN, Fricke R, van der Laan R (2017) Catalog of fishes (California Academy of Sciences, San Francisco). http://researcharchive.calacademy.org/research/ichthyology/catalog/ fishcatmain.asp [accessed 5 June 2017]
- García-Alzate CA, Urbano-Bonilla A, Taphorn DC (2017) A new species of *Hyphessobrycon* (Characiformes, Characidae) from the upper Guaviare River, Orinoco River Basin, Colombia. ZooKeys 668: 123–138. https://doi.org/10.3897/zookeys.668.11489
- González MF, Díaz-Pulido A, Mesa LM, Corzo G, Portocarrero-Aya M, Lasso C, Chaves ME, Santamaría M (Eds) (2015) Catálogo de biodiversidad de la región orinoquense. Volumen 1.

Serie Planeación ambiental para la conservación de la biodiversidad en áreas operativas de Ecopetrol. Proyecto Planeación ambiental para la conservación de la biodiversidad en las áreas operativas de Ecopetrol. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt - Ecopetrol S.A. Bogotá D.C., Colombia, 408 pp.

- Hsieh TC, Ma KH, Chao A (2016) iNEXT: iNterpolation and EXTrapolation for species diversity. R package version 2.0.12. http://chao.stat.nthu.edu.tw/blog/software-download/
- IGAC-Instituto Geográfico Agustín Codazi (1999) Paisajes fisiográficos de Orinoquia-Amazonia (ORAM) Colombia. Análisis Geográficos, Colombia, 218 pp.
- Lasso C, Usma-Oviedo JS, Trujillo F, Rial A (Eds) (2010) Biodiversidad de la cuenca del Orinoco: bases científicas para la identificación de áreas prioritarias para la conservación y uso sostenible de la biodiversidad. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, WWF Colombia, Fundación Omacha, Fundación La Salle, Instituto de Estudios de la Orinoquia (Universidad Nacional de Colombia). Bogotá, D. C., Colombia, 609 pp.
- Lasso C, Machado-Allison A, Taphorn DC (2016) Fishes and aquatic habitats of the Orinoco River Basin: diversity and conservation. Journal of fish biology 89(1): 174–191. https:// doi.org/10.1111/jfb.13010
- Machado-Allison A, Lasso C, Usma-Oviedo J, Sánchez-Duarte P, Lasso-Alcalá O (2010) Peces. In: Lasso C, Usma-Oviedo J, Trujillo F, Rial A (Eds) Biodiversidad de la Cuenca del Orinoco: Bases científicas para la identificación de áreas prioritarias para la conservación y uso sostenible de la biodiversidad. Instituto de Investigaciones de Recursos Biológicos Alexander von Humboldt, WWF Colombia, Fundación Omacha, Fundación la Salle, Institutos de Estudios de la Orinoquia de la Universidad Nacional de Colombia. Bogotá, D.C., Colombia, 217–255.
- Maldonado-Ocampo JA, Vari RP, Usma-Oviedo JS (2008) Checklist of the freshwater fishes of Colombia. Biota Colombiana 9: 143–237.
- Maldonado-Ocampo JA, Urbano-Bonilla A, Preciado JV, Bogotá-Gregory JD (2013) Peces de la cuenca del río Pauto, Orinoquia Colombiana. Biota Colombiana 14: 114–137.
- Mojica JI, Usma-Oviedo JS, Álvarez-León R, Lasso C (Eds) (2012) Libro rojo de peces dulceacuícolas de Colombia. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, Instituto de Ciencias Naturales de la Universidad Nacional de Colombia, WWF Colombia, Universidad de Manizales. Bogotá, D.C., 164 pp.
- Oliveira C, Avelino G, Abe K, Mariguela T, Benine R, Ortí G, Vari RP, Corrêa-Castro RM (2011) Phylogenetic relationships within the speciose family Characidae (Teleostei: Ostariophysi: Characiformes) based on multilocus analysis and extensive ingroup sampling. BMC evolutionary biology 11(275). https://doi.org/10.1186/1471-2148-11-275
- Team RC (2017) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna. https://www.r-project.org/
- Ramírez-Gil H, Ortega-Lara A, Ajiaco-Martínez RE, Pineda-Arguello I (2011) Ictiofauna del río Orotoy: distribución e importancia. Villavicencio, Colombia. Posgrados en Gestión ambiental Sostenible. Universidad de los Llanos, Villavicencio, 260 pp.
- Reis RE, Kullander SO, Ferraris JC (Eds) (2003) Checklist of the Freshwater Fishes of South and Central America. Edipucrs, Porto Alegre, 729 pp.

- Reis RE, Albert JS, Di Dario F, Mincarone MM, Petry P, Rocha LA (2016) Fish biodiversity and conservation in South America. Journal of Fish Biology 89: 12–47. http://dx.doi. org/10.1111/jfb.13016
- Ribeiro MD, Teresa FB, Casatti L (2016) Use of functional traits to assess changes in stream fish assemblages across a habitat gradient. Neotropical Ichthyology 14(1): e140185. http:// dx.doi.org/10.1590/1982-0224-20140185
- Rodríguez MA, Winemiller KO, Lewis Jr WM, Taphorn D (2007) The freshwater habitats, fishes, and fisheries of the Orinoco River basin. Aquatic Ecosystem Health & Management 10: 140–152. http://dx.doi.org/10.1080/14634980701350686
- Rybicki J, Hanski I (2013) Species-area relationships and extinctions caused by habitat loss and fragmentation. Ecology Letters 16: 27–38. https://doi.org/10.1111/ele.12065
- Sabaj-Pérez MH (Ed.) (2016) Standard symbolic codes for institutional resource collections in herpetology and ichthyology: an Online Reference. Version 6.16. American Society of Ichthyologists and Herpetologists, Washington, D.C. http://www.asih.org/ [accessed 24 June 2017]
- Teresa FB, Casatti L, Cianciaruso MV (2015) Functional differentiation between fish assemblages from forested and deforested streams. Neotropical Ichthyology 13: 361–370. http:// dx.doi.org/10.1590/1982-0224-20130229
- Thomaz AT, Arcila D, Ortí G, Malabarba LR (2015) Molecular phylogeny of the subfamily Stevardiinae Gill, 1858 (Characiformes: Characidae): classification and the evolution of reproductive traits. BMC Evolutionary Biology 15(1). https://doi.org/10.1186/s12862-015-0403-4
- Toussaint A, Charpin N, Brosse S, Villéger S (2016) Global functional diversity of freshwater fish is concentrated in the Neotropics while functional vulnerability is widespread. Nature Scientific Reports 6 (22125). https://doi.org/10.1038/srep22125
- Trujillo A, Suárez C, Usma-Oviedo JS, Trujillo F, Bravo A, Córdoba M, Villa-Navarro F, Yara C, Ayala LT, Acosta A, Alfaro JP, Castillo L, Garavito J, Urbano-Bonilla A, Zamudio J, Peña J, Vergel D, Combariza R, Cuéllar J (2011) Ecosistemas estratégicos del Casanare: áreas de alto valor de conservación. In: Usma-Oviedo JS, Trujillo F (Eds) Biodiversidad del Casanare: Ecosistemas Estratégicos del Departamento. Gobernación de Casanare WWF Colombia. Capítulo, Bogotá D.C., 24–49.
- Urbano-Bonilla A, Zamudio J, Maldonado-Ocampo JA, Bogotá-Grégory JD, Cortés-Millán G, López L (2009) Peces del piedemonte del departamento de Casanare, Colombia. Biota Colombiana 10: 149–162.
- Urbano-Bonilla A, Maldonado-Ocampo JA (2013) Peces. In: Mora-Fernández C, Peñuela Recio L (Eds) Evaluación de la Salud Ecosistémica de las sabanas inundables asociadas a la cuenca del río Pauto. Yoluka ONG, Fundación de Investigación en Biodiversidad y Conservación, Fundación Horizonte Verde, Ecopetrol S.A. Bogotá D.C., 277–336.
- Urbano-Bonilla A, Prada-Pedreros S, Zapata A, Barrera-Cataño J, Moreno-Cárdenas C (2014) Composición y riqueza íctica en quebradas y ríos del piedemonte de la cuenca del río Cusiana (Orinoquia colombiana). Biota Colombiana 15: 31–48.
- Urbano-Bonilla A, Zamudio J, Maldonado-Ocampo JA (2016) Ecological aspects of *Lebiasina* erythrinoides (Characiformes: Lebiasinidae) From an Andean piedmont stream in Colombia. Universitas Scientiarum 21: 83–97. http://dx.doi.org/10.11144/Javeriana.SC21-1.eaol

- Usma-Oviedo JS, Villa-Navarro F, Lasso C, Castro F, Zúńiga-Upegui PT, Cipamocha C, Orte-
- ga-Lara A, Ajiaco RE, Ramírez-Gil H, Jiménez LF, Maldonado-Ocampo JA, Muñoz J, Suárez JT (2013) Peces dulce acuícolas de Colombia. In: Zapata LA, Usma JS (Eds) Guía de las Especies Migratorias de la Biodiversidad en Colombia. Peces. Vol. 2. Ministerio de Ambiente y Desarrollo Sostenible/WWF-Colombia. Bogotá, D.C. Colombia, 79–128.
- Usma-Oviedo JS, Maldonado-Ocampo JA, Villa-Navarro FA, Ortega-Lara A, Taphorn D, Urbano-Bonilla A, Zamudio JE, DoNascimiento C (2016) Peces de la cuenca del río Meta. In: Trujillo F, Antelo R, Usma-Oviedo JS (Eds) Biodiversidad de la cuenca baja y media del río Meta. Fundación Omacha, Fundación Palmarito, WWF, Bogotá, 104–119.
- Vanegas-Ríos JA, Urbano-Bonilla A, Azpelicueta M de las M (2015) *Chrysobrycon guahibo*, a new species from the Orinoco River basin, with a distribution expansion of the genus (Teleostei: Characidae). Ichthyological Explorations of the Freshwaters 26: 171–182.
- Villa-Navarro F, Urbano-Bonilla A, Ortega-Lara A, Taphorn D, Usma-Oviedo JS (2011) Peces del Casanare. In: Usma JS, Trujillo F (Eds) Biodiversidad del departamento del Casanare, identificación de ecosistemas estratégicos. Gobernación del Casanare, WWF, Bogotá D.C., 120–137.
- Wieczorek J, Bloom D, Guralnick R, Blum S, Döring M, Giovanni R, Robertson T, Vieglais D (2012) Darwin Core: an evolving community-developed biodiversity data standard. PloS One 7(1): e29715. https://doi.org/10.1371/journal.pone.0029715
- Zamudio J, Urbano-Bonilla A, Maldonado-Ocampo JA, Bogotá-Gregory JD, Cortés-Millán G (2008) Hábitos alimentarios de diez especies de peces del departamento del Casanare, Colombia. Dahlia 10: 43–56.

RESEARCH ARTICLE



# First records of the uristid lysianassoids from Korean waters: redescription of Anonyx abei Takekawa & Ishimaru, 2001 and description of Anonyx exilipes sp. n. (Crustacea, Amphipoda, Uristidae)

Tae Won Jung<sup>1</sup>, Charles Oliver Coleman<sup>1</sup>, Ji Hyung Kim<sup>2</sup>, Seong Myeong Yoon<sup>3</sup>

Museum für Naturkunde Berlin, 10115 Berlin, Germany 2 Infectious Disease Research Center, Korea Research Institute of Bioscience and Biotechnology, Daejeon 34141, South Korea 3 Department of Biology, Chosun University, Gwangju 61452, South Korea

Corresponding author: Seong Myeong Yoon (smyun@chosun.ac.kr)

Academic editor: A. Myers   Received 2 November 2017   Accepted 21 December 2018   F	Published 31 January 2018

**Citation:** Jung TW, Coleman CO, Kim JH, Yoon SM (2018) First records of the uristid lysianassoids from Korean waters: redescription of *Anonyx abei* Takekawa & Ishimaru, 2001 and description of *Anonyx exilipes* sp. n. (Crustacea, Amphipoda, Uristidae). ZooKeys 733: 99–117. https://doi.org/10.3897/zookeys.733.22021

#### Abstract

The uristid lysianassoids are reported for the first time from Korean waters with a redescription of *Anonyx abei* Takekawa & Ishimaru, 2001 and the description of *Anonyx exilipes* **sp. n.** *Anonyx abei* is characterized by a distinctively small projection of the posterodistal corner on epimeron 3, different from all species of the *Anonyx nugax* group which share a constriction at the point of insertion of a distal seta on the inner ramus of uropod 2. *Anonyx exilipes* **sp. n.** is included in the *Anonyx laticoxae* group characterized by the unconstricted inner ramus of uropod 2. This new species is distinguished from other *A. laticoxae* group species by the longer and more slender carpus and propodus of pereopod 6, and the non-lobate merus of pereopod 7.

#### Keywords

Amphipods, Anonyx abei, Anonyx exilipes sp. n., Korea, taxonomy, uristids

#### Introduction

The family Uristidae Hurley, 1963 is a widespread and large group of lysianassoid amphipods containing more than 180 species in 25 well-defined genera (Lowry and Kilgallen 2014, WoRMS 2017). This family is characterized by the modified mouth-parts in most species such that the apical setae of the outer lobe of maxilla 1 show a 7/4 arrangement, the mandibular incisor forms a curved blade, the molar is modified into a setose tongue, and the inner lobe of maxilla 2 is significantly shorter than the outer lobe (Lowry and Stoddart 1989, 1997, Lowry and Kilgallen 2014).

Anonyx Krøyer, 1838 is one of the largest genera of uristids constituted of about 50 species described from the arctic-boreal region (Takekawa and Ishimaru 2001, Lowry and Kilgallen 2014, WoRMS 2017). Steele (1979, 1982, 1983, 1986, 1989, 1991) studied on this genus extensively and divided it into five informal subgroups based on the states of a constriction at the inner ramus on uropod 2: 1) the A. laticoxae group: ramus without a constriction at the insertion of distal robust seta, which equal to or slightly longer than proximal seta; 2) the A. validus group: uropod 2 or inner ramus expanded laterally, lacking a constriction, setae small or lacking; 3) the A. nugax group: inner ramus with a constriction at the point of insertion of a distal seta, which longer than proximal setae; 4) the A. compactus group: inner ramus constricted beyond the point of insertion of the distal seta which much longer than proximal setae; and 5) the A. bispinosus group: inner ramus completely constricted beyond the point of insertion of the distal seta, and the proximal portion of the inner ramus laterally flattened. Until now, it is not certain if these taxonomic groupings of Steele are reflecting the phylogeny. Nevertheless, these groups are useful for identification of the Anonyx species, because the states of constriction at the inner ramus on uropod 2 can easily be observed.

In spite of the species abundance and wide range of distribution, the taxonomic study on lysianassoids is insufficient in Korea and only eight species have been reported: *Aroui minusetosus* Jung, Coleman & Yoon, 2017; *Lepidepecreum vitjazi* Gurjanova, 1962; *Orchomenella japonica* Gurjanova, 1962; *Orchomenella obtusa* (GO Sars, 1891); *Orchomenella paucisetigera* Jung, Yi, Coleman & Yoon, 2017; *Orchomenella rugosa* Jung, Yi, Coleman & Yoon, 2017; *Pseudorchomene boreoplebs* Jung, Coleman & Yoon, 2017; and *Socarnes tongyeongensis* Kim & Hendrycks, 2013 (The Korean Society of Systematic Zoology 1997, Jung and Kim 2008, Kim and Hendrycks 2013, Jung et al. 2015, 2017a, 2017b, 2017c). However, none of these are members of the Uristidae. This is the first record of the family Uristidae from Korean waters.

#### Materials and methods

Collected specimens were initially fixed in 80% ethyl alcohol in the field and then preserved in 95% ethyl alcohol after sorting in the laboratory. Specimens were stained with lignin pink before dissection. Their appendages were dissected in petri dishes or on excavated microscopic slides filled with glycerol using forceps and needles under

a stereomicroscope (Leica M205). They were mounted onto temporary slides using glycerol-ethanol mixed solution or permanent slides using polyvinyl lactophenol solution. For making illustrations, pencil drawings were performed under a light microscope (Leica DMLB) with the aid of a drawing tube. Drawings were scanned, inked digitally and arranged to plates using the methods described in Coleman (2003, 2009). Examined materials are deposited at the National Institute of Biological Resources (**NIBR**) of Korea.

#### Systematic accounts

#### Order Amphipoda Latreille, 1816 Superfamily Lysianassoidea Dana, 1849

#### Family Uristidae Hurley, 1963

Korean name: Na-do-gin-pal-yeop-sae-u-gwa, new

#### Genus Anonyx Krøyer, 1838

Korean name: Na-do-gin-pal-yeop-sae-u-sok, new

#### Anonyx abei Takekawa & Ishimaru, 2001

Figs 1–4 Korean name: Na-do-gin-pal-yeop-sae-u, new

Anonyx abei Takekawa & Ishimaru, 2001: 410, figs 6-10.

**Material examined.** One male (9.3 mm) and one female (7.8 mm), NI-BRV0000807162, Korea: Jeju-do, Beom Is., 33°12.9945N 126°32.215E, depth 66 m, 1 Nov 2016, collected by a light trap.

**Diagnosis.** Gnathopod 1 subchelate; basis weakly setose anteriorly; propodus posterior margin forming weak lobe together palm posterodistally, palm serrated, defined by one pair of elongate robust setae; dactylus with strong protrusion on inner margin. Gnathopod 2 minutely chelate; propodus subquadrate, with nine robust setae anterodistally, posterodistal corner produced distally with two robust setae, palm short, with small cavity; dactylus anchored at middle of distal margin on propodus, inner margin denticulate. Epimeron 2 posteroventral corner a little produced. Uropod 2 inner and outer rami each with a constriction at insertion point of distal elongate seta on dorsal surface.

**Description of male.** *Head* (Fig. 1B). Lateral cephalic lobes expanded anteriorly, subtriangular, apex rounded; eye large, pyriform, occupying most of anterior part of head, composed of numerous small ommatidia.



**Figure 1.** *Anonyx abei* Takekawa & Ishimaru, 2001, male, NIBRV0000807162, 9.3 mm. **A** habitus **B** head **C** antenna 1, medial **D** antenna 2 **E** right mandible **F** left mandible. Scale bars: 0.2 mm (**B–E**), 0.5 mm (**A**).



**Figure 2.** *Anonyx abei* Takekawa & Ishimaru, 2001, male, NIBRV0000807162, 9.3 mm. **A** lower lip **B** maxilla 1 **C** maxilla 1 inner lobe **D** maxilla 2 **E** maxilliped **F** maxilliped inner lobe **G** gnathopod 1 **H** gnathopod 1 palm and dactylus, enlarged I coxa 1. Scale bars: 0.05 mm (**H**), 0.1 mm (**A–F**), 0.2 mm (**G, I**).

Antenna 1 (Fig. 1C) distinctly shorter than antenna 2; peduncle 1<sup>st</sup> article largest, weakly expanded; accessory flagellum composed of five articles, 1<sup>st</sup> article longest, with five robust setae on posterior margin; flagellum 1<sup>st</sup> article distinctly elongate, with one robust seta at posterodistal corner, 2<sup>nd</sup> article with one pair of robust setae at posterodistal corner, calceoli present from 3<sup>rd</sup> article.

Antenna 2 (Fig. 1A, D) elongate,  $0.4 \times$  as long as body; gland cone developed but apex blunt; peduncle 4<sup>th</sup>, 5<sup>th</sup> articles expanded distally, setose on anterior margin; flagellum composed of 34 articles; calceoli present anterodistally.

*Mandible* (Fig. 1E, F) incisor smooth, but bearing blunt denticles on both sides; lacinia mobilis present on left side only, narrowly cylindrical (finger-like), slender; three small raker setae and a patch of short setules present between raker setae and molar processes; molar process not triturative, flap-shaped, densely pubescent, lateral setigerous crest present; palp composed of three articles, attached nearly at level of molar process;  $2^{nd}$  article longest, with an oblique row of ten setae distally;  $3^{rd}$  article falcate,  $0.8 \times as \log as 2^{nd}$  article, inner margin lined with setae, apex with four setae.

Lower lip (Fig. 2A) densely pubescent; inner lobe indistinct.

*Maxilla 1* (Fig. 2B, C) inner lobe short, subquadrate distally, with two plumose setae on blunt apex; outer lobe with eleven toothed setae in 7/4 arrangement; palp bi-articulate, distal article width steady, slightly curved, with eight robust setae on apical margin.

*Maxilla 2* (Fig. 2D) inner lobe reduced, half as long as outer lobe, narrowing distally, with two rows of simple and plumose setae on mediodistal margin (proximal plumose seta longest); outer lobe also narrowing distally and with two setal rows on mediodistal margin.

*Maxilliped* (Fig. 2E, F) inner lobe subrectangular with one mediodistal row of plumose setae, apex blunt with three nodular setae; outer lobe well developed, subovoid, not beyond palp 3<sup>rd</sup> article, lined with 16 nodular setae on mediodistal margin (all nodular setae small); palp composed of four articles, 1<sup>st</sup> article expanded, 2<sup>nd</sup> article with setae medially, 3<sup>rd</sup> article slender, 4<sup>th</sup> article half as long as article 3, apical seta robust, short.

**Pereon.** Gnathopod 1 (Fig. 2G–I) subchelate; coxa large, subquadrate, slightly expanded anteroventrally, with one small notch at posteroventral corner; basis  $0.7 \times as$  long as coxa, width nearly steady, anterior margin a little lobate distally, weakly setose; ischium moderate, with one small anterior lobe; merus triangular,  $0.4 \times as$  long as basis; carpus half as long as basis, convex anteroproximally, carpal lobe blunt, lined with minute setae; propodus  $0.9 \times as$  long as carpus, gradually diminished distally but forming weak lobe together palm posterodistally, palm distinct, serrated, defined by one pair of elongate robust setae; dactylus falcate, exceeding palm, with strong protrusion on inner margin.

Gnathopod 2 (Fig. 3A–D) slender, minutely chelate; coxa subrectangular, slightly divergent ventrally, with one small notch posteroventrally; basis as long as coxa, curved in midway; ischium elongate,  $0.6 \times$  as long as basis; merus  $0.7 \times$  as long as ischium, with numerous short setae posteriorly, posterodistal corner angular with many elongate setae; carpus  $0.7 \times$  as long as basis, margins and medial surface covered with numerous short setae, with elongate setae anteriodistally (longest seta exceeding pro-

podus), posterior margin distal half also with elongate setae; propodus subquadrate, with nine robust setae anterodistally, posterodistal corner produced distally with two robust setae, palm short, with small cavity; dactylus short, anchored at middle of distal margin on propodus, inner margin denticulate, apex slightly exceeding corner of palm.

*Pereopod 3* (Fig. 3E, F) coxa subrectangular,  $0.4 \times as$  wide as long, with one small notch posteroventrally; basis  $0.6 \times as$  long as coxa, somewhat expanded posterodistally; ischium moderate size, with one small anterior lobe; merus expanded anteriorly, slightly tipped anterodistally; carpus  $0.6 \times as$  long as merus, not expanded; propodus  $1.5 \times as$  long as carpus, lined with simple setae on posterior margin, with one hooked locking seta posterodistally; dactylus falcate, elongate,  $0.4 \times as$  long as propodus, unguis weak.

*Pereopod 4* (Fig. 3G, H) coxa deeper than wide, expanded posteroventrally; other articles nearly similar with those of pereopod 3.

*Pereopod 5* (Fig. 4A) coxa large, subquadrate,  $1.2 \times$  as wide as long, equilobate; basis subovoid, anterior margin rounded, lined with robust setae, with one pair of robust setae anterodistally (one seta elongate), posterior lobe well developed, more expanded proximally, margin somewhat flattened, crenulate, expanded posterodistal corner exceeding ischium; ischium to carpus lined with elongate slender setae and short setae anteriorly; merus expanded posteriorly; carpus subrectangular, narrowing distally,  $1.2 \times$  as long as merus; propodus linear,  $1.3 \times$  as long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times$  as long as propodus, unguis weak.

*Pereopod 6* (Fig. 4B) longer than pereopod 5; coxa subrectangular, smaller than that of pereopod 5, bilobate, anterior lobe small, posterior lobe more expanded posteroventrally; basis subovoid,  $0.7 \times$  as wide as long,  $1.1 \times$  as long as that of pereopod 5, anterior margin lined with robust setae on distal 2/3 margin, with one pair of robust setae anterodistally (one seta elongate), posterior lobe well developed, margin somewhat flattened, crenulate, expanded posterodistal corner not exceeding ischium; elongate slender setae present from ischium to merus anteriorly; merus subrectangular,  $0.4 \times$  as long as basis, a little expanded posteriorly, tipped posterodistally; carpus rectangular, not lobate,  $1.3 \times$  as long as merus, a little curved and slightly diminished distally; propodus slender, as long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times$  as long as propodus, unguis weak.

*Pereopod 7* (Fig. 4C, D) longer than pereopod 6; coxa unilobate, as large as that of pereopod 6, expanded posteroventrally; basis  $1.2 \times as$  wide and  $1.1 \times as$  long as that of pereopod 6, anterior margin slightly concaved at the middle, with robust setae on distal 3/4 margin, posterior lobe well developed, margin rounded, not flattened than those of pereopods 6–7; slender setae absent in ischium and merus; merus rectangular, not lobate,  $0.3 \times as$  long as basis, weakly produced posterodistally; carpus also rectangular,  $1.2 \times as$  long as merus; propodus slender,  $1.1 \times as$  long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times as$  long as propodus, unguis weak.

**Pleon.** Epimeron 1 weakly produced anteroventrally, rounded posteroventrally. Epimeron 2 larger than epimeron 1, also produced anteroventrally, slightly convex ventrally, posteroventral corner a little produced. Epimeron 3 largest, regularly rounded posteroventrally, posteroventral corner produced backwards. Urosomite 1 with deep dorsal depression and distal carina weak (Fig. 4E).



Figure 3. Anonyx abei Takekawa & Ishimaru, 2001, male, NIBRV0000807162, 9.3 mm. A gnathopod 2
B gnathopod 2 plam and dactylus C gnathopod 2 carpus to dactylus, medial D coxa 2 E pereopod 3
F pereopod 3 locking seta G pereopod 4 H pereopod 4 locking seta. Scale bars: 0.05 mm (B, F, H), 0.2 mm (A, C-E, G).



**Figure 4.** *Anonyx abei* Takekawa & Ishimaru, 2001, male, NIBRV0000807162, 9.3 mm. **A** pereopod 5 **B** pereopod 6 **C** pereopod 7 **D** pereopod 7 carpus to dactylus **E** pleonal epimera 1–3, lateral **F** uropod 1 **G** uropod 2 **H** uropod 2 outer ramus **I** uropod 2 inner ramus **J** uropod 3 **K** telson. Scale bars: 0.2mm (**A-D, F–K**), 0.5 mm (**E**).

*Uropod 1* (Fig. 4F) longest; peduncle  $1.2 \times as$  long as inner ramus, with eleven robust setae on dorsolateral margin and seven elongate robust setae on dorsomedial margin; rami subequal to each other; inner ramus with four dorsomedial and two dorsolateral robust setae (distal setae on both sides more robust and bearing wrinkly surfaces); outer ramus with three dorsolateral robust setae (distal two setae more robust and bearing wrinkly surfaces).

*Uropod 2* (Fig. 4G–I)  $0.7 \times as$  long as uropod 1; peduncle as long as inner ramus, with six robust setae on dorsolateral margin and three robust setae on dorsomedial margin; inner ramus  $1.1 \times as$  long as outer ramus, with one dorsolateral and two dorsomedial setae, with one constriction at insertion point of distal elongate seta on dorsal surface; outer ramus with three dorsolateral robust setae (with one constriction at insertion point of distal robust setae).

*Uropod 3* (Fig. 4J)  $0.8 \times as$  long as uropod 2; peduncle  $0.7 \times as$  long as inner ramus; both rami with plumose setae on medial margin; outer ramus bi-articulate, distal article  $0.3 \times as$  long as proximal article; inner ramus as long as inner ramus.

*Telson* (Fig. 4K) longer than broad, cleft to about 80%, each lobe with apical notch bearing one pair of robust seta and sensory seta.

**Remarks.** The Anonyx nugax group of Steele (1982) is characterized by the presence of a constriction at the point of insertion of a distal seta which is longer than the proximal setae on the inner ramus of uropod 2. Takekawa and Ishimaru (2001) reported Anonyx abei as a new species from Japanese waters, and they assigned this species to the Anonyx nugax group based on the shape of the inner ramus on uropod 2, as mentioned above. Anonyx abei was differentiated from other 13 species included in the A. nugax group by the distinctively small projection of the posterodistal corner on epimeron 3 (Takekawa and Ishimaru 2001). As a result of the profound morphological examination, our Korean specimens are also show this character state and other characteristics also agree with the original description of Takekawa and Ishimaru (2001). However, there are some minor differences between the Korean and Japanese specimens: 1) the accessory flagellum is composed of five articles in Korean specimens (vs. six articles in Japanese specimens), 2) the large teeth on medial edges of incisors are absent in Korean specimens (vs. two and one tooth on left and right mandibles, respectively in Japanese specimens), and 3) there are eight robust setae on the apical margin of the palp of maxilla 1 (vs. six setae in Japanese specimens).

#### Anonyx exilipes sp. n.

http://zoobank.org/F41912F7-0DA7-435C-A75C-FD6A841EB753 Figs 5–8 Korean name: Gin-da-ri-na-do-gin-pal-yeop-sae-u, new

**Type locality.** Near Daejin Port, Daejin-ri Hyeonnae-myeon Goseong-gun Gangwondo South Korea. The specimens were collected from fishery nets of this port. According to the statements of fishermen, these nets were brought out within a 5 km radius from
Daejin Port and the nets were deployed in about 1–2 km depth (The precise coordinates were uncertain).

Material examined. Holotype: Male (23.0 mm), NIBRIV0000806537, paratypes: two males and two females (18.5 mm–24.9 mm), NIBRIV0000807160; 11 Mar 2016, by TW Jung.

**Etymology.** The composite epithet of the specific name, *exilipes*, is a combination of the Latin *exilis* and *pes*. This name means 'slender foot' referring to the slender shapes of pereopods 6 and 7 compared to those of other species of the *Anonyx laticoxae* group.

**Diagnosis.** Gnathopod 1 basis with setae along entire anterior margin; palm serrated; dactylus without protrusion. Gnathopod 2 propodus half as long as carpus, posterodistal corner produced distally, palm short, with small cavity; dactylus short, apex not exceeding corner of palm. Pereopods 3–4 each propodus with single locking setae posterodistally. Pereopod 6 carpus and propodus elongate, slender. Pereopod 7 merus not lobate. Epimeron 2 posteroventral corner acutely produced backwards. Epimeron 3 regularly rounded posteroventrally, posterior margin produced backwards. Uropod 2 both rami without constrictions. Uropod 3 inner ramus as long as proximal article of outer ramus.

**Description of holotype male.** *Head* (Fig. 5B). Lateral cephalic lobes expanded anteriorly, subtriangular, apex rounded; eye large, pyriform, occupying most of anterior part of head, composed of numerous small ommatidia.

*Antenna 1* (Fig. 5C, D) distinctly shorter than antenna 2; peduncle 1<sup>st</sup> article ovoid, expanded; 2<sup>nd</sup> and 3<sup>rd</sup> articles reduced; accessory flagellum composed of eight articles, 1<sup>st</sup> article longest, dilated distally, lined with several clusters of minute setae on posterior margin; flagellum 1<sup>st</sup> article distinctly elongate, calceoli present from 7<sup>th</sup> article.

Antenna 2 (Fig. 5A, E) elongate,  $0.4 \times$  as long as body; peduncle 4<sup>th</sup>, 5<sup>th</sup> articles convex posteriorly; 4<sup>th</sup> article setose on anterior margin; flagellum composed of 63 articles; calceoli present anterodistally.

Lower lip (Fig. 5F) densely pubescent; inner lobe distinct.

*Mandible* (Fig. 5G, H) incisor smooth but bearing blunt denticles on both sides; lacinia mobilis absent on both sides; nine and eight small raker setae on left and right mandibles respectively; molar process not triturative, flap-shaped, densely pubescent, lateral setigerous crest present; palp composed of three articles, attached nearly at level of molar process, 2<sup>nd</sup> article longest, setose anterodistally, 3<sup>rd</sup> article falcate 0.8 × as long as 2<sup>nd</sup> article, lined with setae on inner margin and apex.

*Maxilla 1* (Fig. 6B, C) inner lobe short, subquadrate distally, with two plumose setae on blunt apex; outer lobe with eleven toothed setae in 7/4 arrangement; palp composed of two articles, distal article slightly dilated and curved distally, with eight robust setae on apical margin.

*Maxilla 2* (Fig. 6D) inner lobe reduced, half as long as outer lobe, narrowing distally, with two rows of simple and plumose setae on mediodistal margin (proximal plumose seta longest); outer lobe also narrowing distally and with two setal rows on mediodistal margin.

*Maxilliped* (Fig. 6A) inner lobe with mediodistal row of plumose setae, apex rounded with three nodular setae; outer lobe well developed, subovoid, not beyond



Figure 5. Anonyx exilipes sp. n. holotype, male, NIBRIV0000806537, 23.0 mm. A habitus B head
C antenna 1, lateral D antenna 1, medial E antenna 2 peduncular articles F lower lip G right mandible
H left mandible. Scale bars: 0.5 mm (C–H), 1.0 mm (B), 2.0 mm (A)



**Figure 6.** Anonyx exilipes sp. n. holotype, male, NIBRIV0000806537, 23.0 mm. **A** maxilliped **B** maxilla 1 **C** maxiila 1 inner lobe **D** maxilla 2 **E** gnathopod 1 **F** gnathopod 1 palm and dactylus. Scale bars: 0.2 mm (**C**, **F**), 0.5 mm (**A**, **B**, **E**).

the palp  $3^{rd}$  article lined with many nodular setae from medial to distal half of lateral margins (all nodular setae small); palp composed of four articles,  $2^{nd}$  article 1.1 × as long as  $1^{st}$  article, with setae medially,  $3^{rd}$  article slightly dilated distally,  $0.7 \times$  as long as  $2^{nd}$  article, covered with minute setae distally and with many elongate setae,  $4^{th}$  article  $0.7 \times$  as long as  $3^{rd}$  article, apical seta robust, short.

**Pereon.** Gnathopod 1 (Fig. 6E, F) subchelate; coxa large, subtrapezoidal, expanded anteroventrally, posteroventral notch nearly weak; basis stout, as long as coxa, anterior margin straight, with setae along entire margin, posterior margin expanded distally, smooth, only with one cluster of setae at distal corner; ischium moderate in size, with one small anterior lobe; merus triangular,  $0.3 \times$  as long as basis, covered with minute setae posteriorly; carpus half as long as basis, convex anteroproximally, carpal lobe weak, apex rounded and covered with minute setae; propodus as long as carpus, gradually diminished distally but forming weak lobe together palm posterodistally, palm distinct, convex, serrated, defined by one pair of elongate robust setae; dactylus falcate, exceeding palm, without protrusion on inner margin, unguis developed.

*Gnathopod 2* (Fig. 7A–B) slender, minutely chelate; coxa subrectangular, slightly divergent ventrally, posteroventral notch nearly weak; basis  $1.1 \times as$  long as coxa, curved at distal 2/3 length; ischium elongate, half as long as basis; merus  $0.8 \times as$  long as ischium, with numerous short setae posteriorly, posterodistal corner angulate with many elongate setae; carpus  $0.6 \times as$  long as basis, anterior margin with three clusters of elongate setae on distal half (longest seta of distal cluster exceeding propodus), carpal lobe flattened, distal half margin also with elongate setae and covered with minute setae; propodus subrectangular, half as long as carpus, margins convex, lateral surface densely covered with setae, posterodistal corner produced distally, palm short, with small cavity; dactylus short, anchored at middle of distal margin on propodus, apex not exceeding corner of palm.

*Pereopod 3* (Fig. 7C, D) coxa subrectangular, half as wide as long, posteroventral notch rather weak; basis  $0.6 \times$  as long as coxa, anterior margin straight, with eleven setae regularly, posterior margin expanded distally; ischium moderate in size, with one small anterior lobe; merus  $0.8 \times$  as long as basis, expanded anteriorly, slightly produced anterodistally; carpus  $0.6 \times$  as long as merus, not expanded; propodus  $1.7 \times$  as long as carpus, lined with paired setae on posterior margin, with one locking seta posterodistally; dactylus falcate, elongate,  $0.4 \times$  as long as propodus, unguis weak.

*Pereopod 4* (Fig. 7E) coxa deeper than wide, expanded posteroventrally; other articles nearly similar with those of pereopod 3.

*Pereopod 5* (Fig. 8A) coxa large, subrectangular,  $1.2 \times as$  wider as long, equilobate; basis subovoid, anterior margin rounded, lined with robust setae, with one pair of robust setae anterodistally (one seta elongate), posterior lobe well developed, more expanded proximally, margin somewhat flattened, crenulate, expanded posterodistal corner not exceeding ischium; ischium to carpus lined with elongate slender setae and short setae anteriorly; merus posterior lobe expanded distally; carpus subrectangular,  $1.3 \times as$  long as merus, posterior margin slightly swollen in midway; propodus linear,  $1.1 \times as$  long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times as$  long as propodus, unguis weak.

*Pereopod 6* (Fig. 8B) longest; coxa subrectangular, smaller than that of pereopod 5, bilobate, anterior lobe small, posterior lobe more expanded posteroventrally; basis subovoid, as wide and  $1.3 \times as$  long as that of pereopod 5, anterior margin lined with robust setae regularly, posterior lobe well developed, margin somewhat flattened, crenulate, expanded posterodistal corner not angulate, not reaching distal end of ischium; elongate slender setae present from ischium to merus anteriorly; merus subrectangular,  $0.4 \times as$  long as basis, slightly expanded posteriorly, weakly produced posterodistally; carpus rectangular, not lobate,  $1.6 \times as$  long as merus, a little curved and slightly diminished distally; propodus slender, linear, as long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times as$  long as propodus, unguis weak.

*Pereopod 7* (Fig. 8C)  $0.9 \times as$  long as pereopod 6; coxa unilobate, as large as that of pereopod 6, expanded posteroventrally; basis  $1.1 \times as$  wide and  $1.1 \times as$  long as that of pereopod 6, anterior margin slightly concaved at the middle, lined with robust setae, posterior lobe well developed, margin rounded, not flattened than those of pereopods 6–7; slender setae absent in ischium and merus; merus rectangular, not lobate,  $0.3 \times as$  long as basis, weakly produced posterodistally; carpus also not lobate,  $1.6 \times as$  long as merus; propodus slender, linear,  $1.1 \times as$  long as carpus, lined with robust setae anteriorly, with one pair of locking setae; dactylus falcate, elongate,  $0.4 \times as$  long as propodus, unguis weak.

**Pleon.** Epimeron 1 (Fig. 8D) weakly produced anteroventrally, rounded posteroventrally. Epimeron 2 (Fig. 8E) slightly larger than epimeron 1, also produced anteroventrally, slightly convex ventrally, posteroventral corner acutely produced backwards. Epimeron 3 (Fig. 8F) largest, regularly rounded posteroventrally, posterior margin produced backwards. Urosomite 1 with deep dorsal depression and distal carina weak (Fig. 5A).

*Uropod 1* (Fig. 8G) longest; peduncle  $1.3 \times as$  long as inner ramus, with twelve robust setae on dorsolateral margin and six elongate robust setae on dorsomedial margin; rami subequal to each other; inner ramus with five dorsomedial and four dorsolateral robust setae; outer ramus with five dorsolateral setae and one dorsomedial seta.

*Uropod 2* (Fig. 8H)  $0.8 \times as$  long as uropod 1; peduncle as long as inner ramus, with eight robust setae on dorsolateral margin and three robust setae on dorsomedial margin; both rami without constriction; inner ramus with two dorsolateral and five dorsomedial setae; outer ramus as long as inner ramus, with five dorsolateral robust setae.

*Uropod 3* (Fig. 8I)  $0.9 \times as$  long as uropod 2; peduncle half as long as inner ramus; both rami with plumose setae on medial margin; outer ramus bi-articulate, distal article  $0.2 \times as$  long as proximal article; inner ramus  $0.9 \times as$  long as proximal article of outer ramus.

*Telson* (Fig. 8J) longer than broad, cleft to about 80%, each lobe with apical notch bearing one pair of robust seta and sensory seta, three or four robust setae and one pair of sensory setae dorsolaterally.

**Remarks.** Steele (1986) divided the genus *Anonyx* into five subgroups according to the shapes of uropod 2. Among them, the *Anonyx laticoxae* group is characterized by sharing of the following features: uropod 2 is narrow, its inner ramus unconstricted,



**Figure 7.** *Anonyx exilipes* sp. n. holotype, male, NIBRIV0000806537, 23.0 mm. **A** gnathopod 2 **B** gnathopod 2 propodus and dactylus, setae omitted **C** pereopod 3 **D** pereopod 3 locking seta and dactylus **E** pereopod 4. Scale bars: 0.2 mm (**B**, **D**), 1.0 mm (**A**, **C**, **E**).



**Figure 8.** *Anonyx exilipes* sp. n. holotype, male, NIBRIV0000806537, 23.0 mm. **A** percopod 5 **B** percopod 6 **C** percopod 7 **D–F** pleonal epimera 1–3 **G** uropod 1 **H** uropod 2 **I** uropod 3 **J** telson. Scale bars: 0.5 mm (**D–J**), 1.0 mm (**A–C**).

and with the distal seta equal to or only slightly longer than the proximal setae. This new species also has this character states and can be included in the *A. laticoxae* group. Moreover, *Anonyx exilipes* sp. n. shares several characteristic features with *Anonyx laticoxae* Gurjanova, 1962 such as similarly produced pleonal epimera, the similar expansions of coxae 1–4, and pereopods 3–4 having single locking setae on their propodus. However, *Anonyx exilipes* sp. n. differs from *A. laticoxae* by the different character states of the carpus and propodus of pereopod 6, which are longer and more slender, and the merus of pereopod 7, which is not lobate in the new species.

#### Acknowledgement

This study was supported by the National Institute of Biological Resources of Korea as a part of the 'Survey of indigenous biological resources of Korea (NIBR NO. 2017-02-001)'.

#### References.

- Coleman CO (2003) "Digital inking": How to make perfect line drawings on computers. Organism, Diversity and Evolution, Electronic Supplement 14: 1–14.
- Coleman CO (2009) Drawing setae the digital way. Zoosystematics and Evolution 85(2): 305–310. https://doi.org/10.1002/zoos.200900008
- Dana JD (1849) Synopsis of the genera of Gammaracea. American Journal of Science and Arts, Series 2, 8: 135–140.
- Gurjanova E (1962) Bokoplavy severnoi chasti Tixogo Okeana (Amphipoda-Gammaridea) chast' 1. Opredeliteli po Faune SSSR 74: 1–440.
- Hurley DE (1963) Amphipoda of the family Lysianassidae from the west coast of North and Central America. Allan Hancock Foundation Publications, Occasional Paper 25: 1–160.
- Jung JW, Kim W (2008) Newly recorded lysianassid species, Orchomene japonicus (Crustacea: Amphipoda) from Korea. Korean Journal of Systematic Zoology 24(1): 139–142. https:// doi.org/10.5635/KJSZ.2008.24.1.139
- Jung TW, Choi HK, Yoon SM (2015) First Record of the Genus Lepidepecreum (Amphipoda: Lysianassidae: Tryphoinae) from Korean Waters. Korean Journal of Environmental Biology 33 (2): 119–125. https://doi.org/10.11626/KJEB.2015.33.2.119
- Jung TW, Coleman CO, Yoon SM (2017a) Aroui minusetosus, a new species of Scopelocheiridae from Korea (Crustacea, Amphipoda, Lysianassoidea). ZooKeys 706: 17–29. https:// doi.org/10.3897/zookeys.706.20007
- Jung TW, Coleman CO, Yoon SM (2017b) Pseudorchomene boreoplebs, a new lysianassid amphipod from Korean waters (Crustacea, Amphipda, Lysianassoidea). Zoosystematics and Evolution 93(2): 343–352. https://doi.org/10.3897/zse.93.20482
- Jung TW, Yi CH, Coleman CO, Yoon SM (2017c) Two new lysianassoid amphipods of the genus Orchomenella (Amphipoda, Lysianassoidea, Lysianassidae, Tryphosinae) from South Korea. Crustaceana 90(13): 1641–1664. https://doi.org/10.1163/15685403-00003740

- Kim Y-H, Hendrycks EA (2013) A new species of *Socarnes* Boeck, 1871 (Crustacea, Amphipoda, Lysianassidae) from Korean waters. ZooKeys 357: 1–10. https://doi.org/10.3897/ zookeys.357.6372
- Krøyer H (1838) Conspectus Crustaceorum Groenlandiae. Naturhistorisk Tidsskrift 2: 249–261.
- Latreille PA (1816) Amphipoda. In: Nouveau Dictionaire d'histoire naturelle, appliquée aux Arts, à l'Agriculture, à l'Économie rurale et domestique, à la Médecine, etc. Par une société de Naturalistes et d'Agriculteurs (2<sup>nd</sup> edn). Volume 1. Deterville, Paris, 467–469.
- Lowry JK, Kilgallen NM (2014) A generic review of the lysianassoid family Uristidae and descriptions of new taxa from Australian waters (Crustacea, Amphipoda, Uristidae). Zootaxa 3867(1): 1–92. https://doi.org/10.11646/zootaxa.3867.1.1
- Lowry JK, Stoddart HE (1989) *Stephonyx*, a new, widespread genus of lysianassoid Amphipoda. Zoologica Scripta 18: 519–525. https://doi.org/10.1111/j.1463-6409.1989.tb00145.x
- Lowry JK, Stoddart HE (1997) Amphipoda Crustacea IV. Families Aristiidae, Cyphocarididae, Endevouridae, Lysianassidae, Scopelocheiridae, Uristidae. Memoirs of the Hourglass Cruises 10: 1–148.
- Sars GO (1890–1895) An account of the Crustacea of Norway with short descriptions and figures of all the species, Vol. I. Amphipoda. Alb. Cammermeyer, Christiana, 711 pp. [240 pls]
- Steele DH (1979) Zoogeography of the genus Anonyx (Crustacea, Amphipoda). Bulletin of the Biological Society of Washington 3: 47–53.
- Steele DH (1982) The genus Anonyx (Crustacea, Amphipoda) in the North Pacific and Arctic Oceans: Anonyx nugax group. Canadian Journal of Zoology 60: 1754–1775. https://doi. org/10.1139/z82-228
- Steele DH (1983) The genus Anonyx (Crustacea, Amphipoda) in the North Pacific and Arctic Oceans: Anonyx validus group. Canadian Journal of Zoology 61: 2921–2931. https://doi. org/10.1139/z83-380
- Steele DH (1986) The genus Anonyx (Crustacea, Amphipoda) in the North Pacific and Arctic Oceans: Anonyx laticoxae group. Canadian Journal of Zoology 64: 2603–2623. https://doi. org/10.1139/z86-380
- Steele DH (1989) The genus Anonyx (Crustacea, Amphipoda) in the North Pacific and Arctic Oceans: Anonyx compactus group. Canadian Journal of Zoology 67: 1945–1954. https:// doi.org/10.1139/z89-278
- Steele DH (1991) The genus Anonyx (Crustacea, Amphipoda) in the North Pacific and Arctic Oceans: Anonyx bispinosus group. Canadian Journal of Zoology 69: 1600–1611. https:// doi.org/10.1139/z91-224
- Takekawa A, Ishimaru S (2001) Two new species of the genus Anonyx (Amphipoda: Gammaridea: Lysianassoidea) from Onagawa Bay, Northeaastern Japan. Zoological Science 18: 405–416. https://doi.org/10.2108/zsj.18.405
- The Korean Society of Systematic Zoology (1997) List of Animals in Korea (excluding insects). Academy Publishing, Seoul, 489 pp.
- WoRMS (2017) World Register of Marine species. Available from: http://www.marinespecies.org [accessed 13 July 2017]

RESEARCH ARTICLE



# An early and mysterious histerid inquiline from Cretaceous Burmese amber (Coleoptera, Histeridae)

Michael S. Caterino<sup>1</sup>, David R. Maddison<sup>2</sup>

l Department of Plant & Environmental Sciences, Clemson University, Clemson, SC 29634, USA 2 Department of Integrative Biology, Oregon State University, Corvallis, OR 97331, USA

Corresponding author: Michael S. Caterino (mcateri@clemson.edu)

Academic editor: J. Klimaszewski   Received 19 December 2017   Accepted 22 January 2018   Published 1 February 201
http://zoobank.org/58E27898-D720-4EDD-9715-D8018EB6D596

**Citation:** Caterino MS, Maddison DR (2018) An early and mysterious histerid inquiline from Cretaceous Burmese amber (Coleoptera, Histeridae). ZooKeys 733: 119–129. https://doi.org/10.3897/zookeys.733.23126

#### Abstract

We describe a new genus and species of Histeridae from Upper Cretaceous Burmese amber, *Amplectister tenax* Caterino & Maddison, **gen. & sp. n.** This species represents the third known Cretaceous histerid, which, like the others, is highly distinct and cannot easily be placed to subfamily. It exhibits prosternal characters in common with Saprininae, but other characters appear inconsistent with this possibility. The abdominal venter is strongly concave, and the hind legs are enlarged and modified for grasping. We hypothesize that this represents the earliest example in Histeridae of modifications for phoresy on social insects.

## Keywords

amber fossil, Upper Cretaceous, phoresy, inquiline

# Introduction

The early diversification of the beetle family Histeridae is poorly understood. Phylogenetic relationships among extant taxa have been difficult to resolve (Caterino and Vogler 2002, McKenna et al. 2015), and the family's fossil record is sparse and poorly documented (Chatzimanolis et al. 2006, Caterino et al. 2015). This uncertainty has hindered studies of ecomorphological evolution, which has followed several distinct and repeated trajectories in the family (Caterino and Vogler 2002). The evolutionary pathways taken by histerid lineages have yielded obligate symbioses with diverse animals, including mammals, birds, and, most spectacularly, with social insects. Many of these obligate inquilines show distinctive suites of morphological characters that facilitate their symbioses (Hölldobler and Wilson 1990), including trichomes, exaggerated development of certain body parts, and defensive modifications. Histeridae as a whole is characterized by a body form and structures that give them an ability to defend themselves against attack, including retraction and protection of appendages. This hints at some early symbioses, although there is little support for this in the existing fossil record.

Recent work has begun to reveal a much greater diversity of early Histeridae than previously suspected. Until quite recently the family's fossil record extended no more than about 40 MYBP (Szwedo and Sontag 2009), but discoveries in Cretaceous Burmese amber have more than doubled this minimum age for the family. Poinar and Brown (2009) described the first of these, *Pantostictus burmanicus*, although the specimens were rather poor and the placement of this species remains unclear. Caterino et al. (2015) described the much better preserved *Cretonthophilus tuberculatus* from the same deposits, hypothesizing placement in Onthophilinae. Here we describe a new genus and species of fossil histerid from the same Burmese amber deposits (with a presumed age of about 99 MYBP, Shi et al. 2012), which offers further insight into the family's earliest history. This species exhibits distinct hallmarks of inquilinism, with an abdominal-metathoracic leg complex clearly adapted for grasping.

## Methods

The original piece of amber (Fig. 1; OSAC lot number OSAC\_AMB0000057) was cut into three pieces, and polished. In one piece is the histerid described here (specimen OSAC\_0002900057); the remaining pieces contain the other synclusions described below. Photographs were taken using Visionary Digital's Passport II imaging system (based on a Canon 6D SLR with 65 mm MP-E 1-5× macro lens). Image stacking was done using Helicon Focus (www.heliconsoft.com). Drawings were penciled by hand, traced on a drawing pad, and 'inked' in Adobe Illustrator. Measurements were taken using a Leica M125 calibrated eyepiece micrometer.

# Systematic paleontology

Family: Histeridae Gyllenhal, 1808 Subfamily: *incertae sedis* 

Amplectister Caterino & Maddison, gen. n. http://zoobank.org/4D931E23-8F6B-4AC6-94C7-3E5229DE3BD2

Type species. Amplectister tenax Caterino & Maddison, sp. n.



**Figure 1.** Photograph of original piece of amber (OSAC\_0002900057) containing holotype before cutting and polishing.

**Diagnosis.** Many features distinguish this extinct genus: overall body form quite elongate and flattened (Figs 2–4); frons laterally carinate and projecting over the antennal insertions (Figs 5–6); pronotum with sinuate posterior margin and broadly arcuate lateral margin that is not aligned with the elytral margin (Fig. 3); elytron with two submarginal epipleural carinae (diverging from the posterior pronotal corner; Fig. 4); abdomen deeply concave (Fig. 2); posterior femora and tibiae enlarged and adapted for grasping (Figs 4, 10).

**Derivation of name.** The genus name (masculine) means 'the hugging Hister', referring to its modifications for grasping, from the Latin *amplexus*.

# Amplectister tenax Caterino & Maddison sp. n.

http://zoobank.org/A06D06E6-52F1-44F0-84A9-CC648422D095

**Type material.** Holotype specimen, of unknown sex; type locality: Northern Myanmar: probably Hukawng Valley, collected in 2016; deposited in Oregon State Arthropod Collection, specimen OSAC\_0002900057. The specimen was purchased by DRM from Yanling Ying in January 2017. Most of his specimens are from the Noije Bum mine or nearby, Kachin State; a few are from around Nam Sakhaw in Sagaing Division (NW of Haungpa); fewer are from elsewhere in other areas in Kachin State.

**Description.** Many body surfaces encrusted with thin off-white granular substance and/or thin film of air; textures and surface sculpture difficult to assess. An oblique planar fracture below the anterior part of the body distorts some observations of ventral anterior structures.

Measurement	mm
Pronotum+elytral (PE) length	1.41
Pronotal length	0.41
Pronotal width	0.98
Elytral length	1.00
Humeral width	1.02
Propygidium length	0.10
Pygidium length	0.24
Head width	0.37
Prosternum length	0.33
Mesoventrite length	0.10
Metaventrite length	0.37
Profemur length	0.35
Protibia length	0.29
Mesofemur length	0.47
Mesotibia length	0.43
Metafemur length	0.73
Metatibia length	0.57

Table 1. Body measurements in millimeters.

Total body (pronotum + elytra) length: 1.41mm; maximum (humeral) width: 1.02mm (for all measurements see Table 1). Body surfaces all apparently finely granulate, matte, possibly finely reticulate, not shiny; dorsal surface lacking obvious punctures; ventral surfaces distinctly punctate on most surfaces.

Frons broad, anteriorly prominent (Figs 5–6); eyes present, large, located on sides of head; longitudinal supraocular ridges projecting anterad eyes, continued mediad by prominent, slightly oblique frontal ridges over antennal and mandibular insertions, frontal ridges possibly continuous medially (obscured); frontoclypeal suture not apparent (probably absent, but obscured); epistoma convex along longitudinal midline; labrum evenly rounded apically, convex, without major setae (though with short setal fringe around edges appressed to mandibles); mandibles apically acute, incisor edges short, neither with secondary tooth, left mandible overlapping right in repose; outer surface of mandibles weakly concave in basal half; head mostly retracted, ventral mouthparts not visible. Antennal scape short, expanded slightly to apex, bearing two elongate setae near apex; pedicel about one-third length of scape, subcylindrical; antennal funicle apparently with 6 more or less transverse antennomeres, gradually widening distad, with antennomere 8 nearly as wide as club; antennal club slightly elongate oval, weakly truncate apically, setose, bearing specialized setose patch on inner apical surface (Fig. 7), outer surface may be lightly sclerotized; antennal annuli not apparent.

Pronotum (Figs 3, 5) rather broad, with deep anterior emargination; sides broadly rounded, distinctly widened from obtuse basal corners, widest about one-fourth from base, converging arcuately to rounded anterior corners; central part of pronotal disk convex, lateral margins depressed to broadly explanate, particularly in anterior corners, edges flattened, slightly reflexed.



Figures 2–5. Photographs of holotype. 2 Ventral view 3 Dorsal view 4 Lateral view 5 Frontal view.

Scutellum present, small, triangular; elytra (Fig. 3) broad, apparently asymmetrical (possibly optical distortion), the right tapered to a narrower apex than left, moderately flattened, lacking distinct striae but with weak serial depressions, posterolateral corners broadly rounded, apices truncate; each elytron with prominent marginal carina delimiting epipleuron extended from humeral corner around posterior corner, though not attaining apical midline; epipleuron (Fig. 4) with secondary carina extending from humeral corner about two-thirds epipleural length, there merging with lower elytral margin; elytral margin not carinate; metathoracic wings present (protruding slightly beneath posterolateral corner of left elytron).



Figures 6–10. Drawings from holotype. 6 Frontal view 7 Prosternum and antennae 8 Prothoracic leg, anterior view 9 Mesothoracic leg, anterior view 10 Metathoracic leg, anterior view.

Propygidium (Fig. 3) exposed, wide, short, bearing numerous stiff setae (this is the only exposed sclerite for which this is true); pygidium subtriangular, with rounded sides and apex, disk depressed with a continuously elevated marginal carina; pygidium slightly opened, but genitalia obscured by air bubbles, sex undeterminable. Prosternum (Figs 2, 7) elevated at middle, anteriorly incised on either side of keel for passage of antennal funicle, with deep rounded depressions along keel and behind prosternal lobe for reception of antennal club; prosternal keel shallowly emarginate at base, keel elevated, with two prominent carinae, parallel from base to near apex, converging slightly above antennal cavities, distinctly depressed between; very short lateral carinae descend from inner anterior edge of profemur to join keel carinae behind antennal cavity; prosternal lobe minimal, forming broad flange delimiting front of antennal cavities, weakly emarginate where mandibles rest. Hypomeron broadly expanded laterally, with oblique longitudinal carina from anterior corner to near outer corner of profemoral insertion.

Mesoventrite (Figs 2, 7) broad, anterior margin sinuate, weakly but distinctly produced at middle; mesometaventral suture apparently impressed (obscured); metaventrite with prominent, oblique postmesocoxal carinae extending from inner corners of mesocoxae to middle of metacoxa; middle of mesoventrite increasingly depressed posterad; laterally, mesepimeron, metepisternum, and metepimeron all distinct, apparently covered with large punctures (somewhat obscured), as is lateral portion of metaventrite.

Abdominal venter (Fig. 2) deeply concave medially; sides of first ventrite elevated behind metacoxae, forming a distally setose lateral flange; subsequent ventrites transversely depressed, with abdomen deeply arched to pygidial apex.

Legs (Figs 8–10): Procoxa moderately and obliquely transverse; protrochanter subquadrate, with inner corner prominent, setose; profemur narrowed to apex, with anterior, upper edge straight, inner edge weakly excavate for reception of protibia, inner posterior edge weakly expanded bearing few prominent setae; protibia narrow at base, widened weakly to apex, with two small apical spurs at inner corner, laterally with weakly bispinose apex, three to four weak denticles bearing small spines basad along margin, inner edge with series of ~8 fine spines; tarsal groove of anterior face of protibia poorly if at all developed; protarsomeres 1-4 short, subequal, bearing pair of ventral spines, apical tarsomere about three times length of tarsomere 4, with two ventral spines along midline, with pair of regular tarsal claws. Mesocoxa rounded; mesofemur narrowed to apex, with few prominent setae along anterior inner edge; mesotibia narrow, with weak apical spurs, outer edge slightly rounded, with single prominent spine at outer apical margin; outer posterior edge weakly grooved to receive tarsus; tarsus as for protarsus. Metacoxae rounded, widely separated; metatrochanter small, obscured, inserted at posterolateral corner of coxa; metafemur broad and thick, with prominent carinae along inner medial, outer medial, and dorsal margins (narrowly triangular in cross-section), inner surface weakly concave for reception of inner edge of metatibia; metatibia broad and flat, inner margin straight and bearing series of fine spines, outer margin rounded, smooth, inner surface with diffuse cluster of stiff setae about onethird from tibial base; metatarsus segmented as for meso- and protarsus, apparently received along apical half of outer edge of medial tibial face.

**Derivation of specific epithet.** The species name means tenacious, referring to its grasp, from the Latin *tenax*.

#### Synclusions

In the same piece of amber as the original specimen were one beetle of the family Eucinetidae (Fig. 1), one mite, and a "stellate hair", presumably of plant origin. The mite specimen was destroyed in cutting and polishing.

# Discussion

Histerid systematics has relied heavily on the form of the prosternum for classification and phylogenetics (Wenzel 1944; Kovarik and Caterino 2016). This new taxon appears very similar to Saprininae in prosternal characters. Modern Saprininae have a nearly identical form of antennal retraction, with an anterior prosternal notch through which the funicle passes, and a deep anterior depression along the side of the prosternal keel for reception of the club. Looking only at these characters this genus could easily be placed in Saprininae, and even close to a genus such as Gnathoncus Jacquelin-Duval. Furthermore, the apparently setose sensory area on the inner surface of the antennal club is strongly suggestive of what in modern Saprininae is termed 'Reichardt's organ', a complex of antennal sensory openings and surfaces (Lackner 2010). Indeed, these prosternal and antennal characters together represent the main morphological synapomorphies of Saprininae (Lackner 2014). However, homology of these characters is not certain, and in numerous other characters Amplectister differs substantially from any modern Saprininae. The projecting frontal margin is not known among modern Saprininae. A much weaker form is seen in *Cretonthophilus*, suggesting this could be a plesiomorphy. The elytral striae in Amplectister are vaguely impressed, but do not show the highly characteristic saprinine set of elytral striae, with the fourth stria arched to the sutural stria. This isn't recognized as a Saprininae synapomorphy by Lackner (2014), but may be. Amplectister exhibits an emarginate prosternal keel, while that in most modern Saprininae extends as a thin laminate projection over the anterior point of the mesoventrite. This has not been formally evaluated in saprinine phylogeny. Finally, Amplectister lacks labral setae, which are universal (though symplesiomorphic) in extant Saprininae. All things considered, it is conceivable that *Amplectister* represents a stem lineage, possessing some but not all apomorphies of extant Saprininae. This possibility merits further exploration and a more quantitative analysis. Deeper examination of Amplectister internal characters through micro-CT scanning (e.g. Perreau and Tafforeau 2011, Riedel et al. 2012) would be particularly informative, as some Saprininae apomorphies are found in the genitalia (Lackner 2014).

Amplectister shows some similarities to the recently described *Cretonthophilus*, sharing short subpyramidal antennal scapes, frontal carinae, concave sides of mandibles, subdepressed body form, elytral and pronotal lateral margins not colinear, and epipleurae carinate, as well as various features of the legs (profemora able to receive protibia, all tibiae flattened, weakly expanded apically, with spines along inner margins). However, our limited understanding of early histerid phylogeny cannot yet distinguish whether any of these could be synapomorphies of the two. Furthermore, significant differences are numerous. The form and manner of reception of the antennal club on the prosternum is very different, with *Cretonthophilus* having a hypomeral cavity far removed from the prosternal lobe. The form of the antennal club itself is also quite different, with that of *Cretonthophilus* showing deep and distinct sutures between the club's three antennomeres. *Cretonthophilus* also has an elongated prosternal lobe, and distinct protibial grooves for reception of its protarsus. These phylogenetically compelling characters suggest that *Cretonthophilus* and *Amplectister* occupy distinct branches of early histerid phylogeny. Regarding possible similarities with *Pantostictus burmanicus*, very little can be said due to the lack of phylogenetically informative characters originally described, or visible in the type specimens, which we have recently examined.

The remarkable ventral modifications of *Amplectister* seem clearly adapted for grasping. Grasping in insects serves several purposes and takes a variety of forms. It seems unlikely that the purpose in *Amplectister* is for grasping prey, since in other insects that grasp prey the raptorial modifications are on anterior portions of the body (e.g., in mantises, mantispids, and various Heteroptera), whereas in *Amplectister* the grasping structures are on posterior regions of the body. Some insects show modifications for grasping various substrates, to resist removal by predators, or to prevent being dislodged (elongate legs and enlarged tarsal claws in lotic systems, for example). As the grasping modifications involve only the hind legs in *Amplectister*, rather than all legs, this also seems unlikely.

The posterior location of these modifications on the body suggest courtship as another possible function, and in many insects males exhibit grasping modifications for retaining hold and position on a mate (e.g. Arnqvist 1989, Miller 2003). In some histerids this often includes some degree of concavity on the venter (Caterino and Tishechkin 2013), though invariably on the metaventrite, and none to the extreme seen in *Amplectister*. However, if such a modification were to facilitate mate-holding, we would expect it to correspond more closely in shape to some part of a similar-shaped female. It is not obvious that it does. Also, it is not clear what purpose the distinctive setose brushes on either side of the abdominal concavity would have in mate-holding, nor what role the large and complicated metathoracic legs would play. Mate-holding as the function of these modifications thus seems unlikely.

We suggest instead that the most likely explanation is related to some form of inquilinism. Histeridae exhibit a variety of symbiotic relationships with other organisms, as obligate inhabitants of bird and mammal nests, as well as guests in ant and termite colonies (Kovarik and Caterino 2016). Many insect inquilines grasp their hosts. In the case of vertebrate hosts, many phoretic and parasitic inquilines show modifications for holding on to the fur, feathers, or other more specific parts of their hosts' bodies. In beetles, the modifications in most such species involve the tarsi (Philips 2011). No such relationships have been described for histerids, but some extant species exhibit chelate tarsi (though not yet directly connected with vertebrate phoresy). Some ant inquilines among Histeridae are known to cling to their host, including the army ant (*Eciton* spp.) guest haeteriines *Nymphister* Reichensperger, which grasps a worker ant's petiole with its mandibles (von Beeren and Tishechkin 2017), and *Pulvinister* Reichensperger, which rides on the underside of major workers' heads (Rettenmeyer 1961). This has also been observed in the chlamydopsine *Chlamydopsis loculosa* Lea, which grasps the thorax of its host (*Rhytidoponera* spp.) with its legs (McMillan 1950). Given the presence of setal projections (possible trichomes) on the abdominal concavity of *Amplectister*, and the unusual leg modifications, a social-insect grasping mechanism seems like a reasonable hypothesis. Although it seems unlikely that any specimens will come to light that will allow us to directly test this, improved resolution of basal histerid phylogeny will permit more detailed phylogenetic assessments of the morphological evolution of all these structures and potentially their relationship to function in early symbioses.

#### Acknowledgments

We would like to thank Mike Ivie for helpful suggestions, and George Poinar for allowing DRM to examine the type specimens of *Pantostictus* from his personal collection. We also thank two anonymous reviewers whose comments improved the manuscript.

#### References

- Arnqvist G (1989) Sexual selection in water strider: the function, nature of selection and heritability of a male grasping apparatus. Oikos 56: 344–350. https://doi.org/10.2307/3565619
- Caterino MS, Wolf-Schwenninger K, Bechly G (2015) *Cretonthophilus tuberculatus*, a remarkable new genus and species of histerid beetle (Coleoptera: Histeridae) from Middle Cretaceous Burmese amber. Zootaxa 4052: 241–245. https://doi.org/10.11646/ zootaxa.4052.2.10
- Caterino MS, Tishechkin AK (2013) A systematic revision of *Operclipygus* Marseul (Histeridae: Histerinae: Exosternini) ZooKeys 271: 1–401. ddoi.org/10.3897/zookeys.271.4062
- Caterino MS, Vogler AP (2002) The phylogeny of the Histeroidea (Staphyliniformia). Cladistics 18(4): 394–415. https://doi.org/10.1016/S0748-3007(02)00002-6
- Chatzimanolis S, Caterino MS, Engel MS (2006) The first fossil of the subfamily Trypanaeinae: A new species of *Trypanaeus* in Dominican amber. The Coleopterists Bulletin 60(4): 333–340. http://www.jstor.org/stable/4009947
- Hölldobler B, Wilson EO (1990) The ants. Cambridge: Harvard University Press, Cambridge, 732 pp. https://doi.org/10.1007/978-3-662-10306-7
- Kovarik PW, Caterino MS (2016) Histeridae. In: Beutel RG, Leschen RAB (Eds) Handbook of Zoology Part 38, Coleoptera, Vol. 1: Morphology and Systematics (2<sup>nd</sup> ed.) Walter de Gruyter, Berlin, 275–314.
- Lackner T (2010) Review of the Palaearctic genera of Saprininae (Coleoptera: Histeridae). Acta Entomologica Musei Nationalis Pragae 50 (Supplement): 1–254.

- Lackner T (2014) Phylogeny of the Saprininae reveals interesting ecological shifts in the history of the subfamily (Coleoptera: Histeridae). Zoological Journal of the Linnean Society 172(3): 521–555. https://doi.org/10.1111/zoj.12182
- McKenna DD, Farrell BD, Caterino MS, Farnum CW, Hawks DC, Maddison DR, Seago A, Short AEZ, Newton AF, Thayer MK (2015) Phylogeny and evolution of Staphyliniformia (and Scarabaeiformia): Litter as a stepping stone for diversification of non-phytophagous beetles. Systematic Entomology 40: 35–60. https://doi.org/10.1111/syen.12093
- McMillan RP (1950) Observations on the jockey beetle (*Chlamydopsis duboulayi*). The Western Australian Naturalist 2: 132–133.
- Miller KB (2003) The phylogeny of diving beetles (Coleoptera: Dytiscidae) and the evolution of sexual conflict. Biological Journal of the Linnean Society, 79: 359–388. https://doi.org/10.1046/j.1095-8312.2003.00195.x
- Perreau M, Tafforeau P (2011) Virtual dissection using phase-contrast X-ray synchrotron microtomography: reducing the gap between fossils and extant species. Systematic Entomology 36: 573–580. https://doi.org/10.1111/j.1365-3113.2011.00573.x
- Philips TK (2011) The evolutionary history and diversification of dung beetles. In: Simmons LW, Ridsdill-Smith TJ (Eds) Ecology and evolution of dung beetles. Blackwell, Oxford, 21–46. https://doi.org/10.1002/9781444342000.ch2
- Poinar G, Brown AE (2009) Pantostictus burmanicus, A new genus and species of Cretaceous beetles (Coleoptera: Hydrophiloidea: Histeridae) in Burmese amber. Proceedings of the Entomological Society of Washington 111(1): 38–46. https://doi.org/10.4289/0013-8797-111.1.38
- Rettenmeyer CW (1961) Arthropods associated with neotropical army ants with a review of the behavior of these ants (Arthropoda; Formicidae: Dorylinae). PhD Thesis Lawrence, KS: University of Kansas, 604 pp.
- Riedel A, Dos Santos Rolo T, Cecilia A, Van De Kamp T (2012) Sayrevilleinae Legalov, a newly recognised subfamily of fossil weevils (Coleoptera, Curculionoidea, Attelabidae) and the use of synchrotron microtomography to examine inclusions in amber. Zoological Journal of the Linnean Society 165: 773–794. https://doi.org/10.1111/j.1096-3642.2012.00825.x
- Shi G, Grimaldi DA, Harlow GE, Wang J, Wang J, Yang M, Lei W, Li Q, Li X (2012) Age constraint on Burmese amber based on U-Pb dating of zircons. Cretaceous Research 37:155– 163. https://doi.org/10.1016/j.cretres.2012.03.014
- Szwedo J, Sontag E (2009) The traps of the "amber trap". How inclusions could trap scientists with enigmas. Denisia, zugleich Kataloge der oberösterreichischen Landesmuseen 26 (Neue Serie 86): 155–169. https://www.zobodat.at/pdf/DENISIA\_0026\_0155-0169.pdf
- von Beeren C, Tishechkin AK (2017) *Nymphister kronaueri* von Beeren & Tishechkin sp. nov., an army ant-associated beetle species (Coleoptera: Histeridae: Haeteriinae) with an exceptional mechanism of phoresy. BMC Zoology 2(3): 1–16. https://doi.org/10.1186/s40850-016-0010-x
- Wenzel RL (1944) On the classification of the histerid beetles. Fieldiana, Zoology 28(2): 51-151.

RESEARCH ARTICLE



# New hairworm (Nematomorpha, Gordiida) species described from the Arizona Madrean Sky Islands

Rachel J. Swanteson-Franz<sup>1</sup>, Destinie A. Marquez<sup>1</sup>, Craig I. Goldstein<sup>2</sup>, Andreas Schmidt-Rhaesa<sup>3</sup>, Matthew G. Bolek<sup>4</sup>, Ben Hanelt<sup>1</sup>

 Center for Evolutionary and Theoretical Immunology, Department of Biology, 163 Castetter Hall, MSC032020, University of New Mexico, Albuquerque, New Mexico 87131-0001, USA 2 Rush Oak Park Hospital, Department of Emergency Medicine, 520 South Maple Avenue, Oak Park, Illinois 60304, USA 3 Zoological Museum and Institute, Biocenter Grindel, Martin-Luther-King-Platz 3, University of Hamburg, 20146 Hamburg, Germany 4 Department of Integrative Biology, 501 Life Sciences West, Oklahoma State University, Stillwater, Oklahoma 74078, USA

Corresponding author: *Ben Hanelt* (bhanelt@unm.edu)

Academic editor: <i>H-P Fagerholm</i>   Received 5 December 2017   Accepted 7 January 2018   Published 1 February 201
http://zoobank.org/DC5CDDD5-74A1-4BF9-BA09-4EC956A57179

**Citation:** Swanteson-Franz RJ, Marquez DA, Goldstein CI, Schmidt-Rhaesa A, Bolek MG, Hanelt B (2018) New hairworm (Nematomorpha, Gordiida) species described from the Arizona Madrean Sky Islands. ZooKeys 733: 131–145. https://doi.org/10.3897/zookeys.733.22798

#### Abstract

Gordiids, or freshwater hairworms, are members of the phylum Nematomorpha that use terrestrial definitive hosts (arthropods) and live as adults in rivers, lakes, or streams. The genus *Paragordius* consists of 18 species, one of which was described from the Nearctic in 1851. More than 150 years later, we are describing a second *Paragordius* species from a unique habitat within the Nearctic; the Madrean Sky Island complex. The Madrean Sky Islands are a series of isolated high mountains in northern Mexico and the southwestern United States (Arizona and New Mexico), and are well known for their high diversity and endemicity. The new species is described based on both molecular data (COI barcoding) and morphological characters of the eggs, larvae, cysts, and adults. Adult females have unique small oblong mounds present on the interior of the trifurcating lobes with randomly dispersed long hairs extending from the furrows between the mounds. Marked genetic differences support observed morphological differences. This species represents the second new hairworm to be described from the Madrean Sky Islands, and it may represent the first endemic hairworm from this biodiversity hotspot.

#### Keywords

Nematomorpha, Gordiid, hairworm, Gordius, Sky Islands, new species

## Introduction

Hairworms are in the phylum Nematomorpha, belonging to one of only 3 entirely parasitic metazoan phyla (Hanelt et al. 2005). Nematomorphs are arthropod parasites with indirect lifecycles, infecting aquatic insect larvae as their paratenic hosts, and orthopterans, coleopterans, or mantids as their definitive hosts (Hanelt et al. 2005). Worms are free living in aquatic environments as adults, where mating and oviposition occur. Larvae subsequently are swallowed by and encyst in suitable paratenic hosts such as midge larva (Hanelt and Janovy 2004a). Terrestrial definitive hosts are infected upon the consumption of infected adult aquatic insects. Upon maturation, the hairworm will alter host behavior so that it becomes water seeking (Biron et al. 2005). Once the definitive host enters the water, the horsehair worms will emerge and begin mating, completing the lifecycle.

Hairworms have been chronically understudied. One estimate suggests that only 14% of species have so far been described (Poinar 2008), and most descriptions have been limited to the Palearctic. Within the Nearctic, one area requiring biodiversity work is the desert Southwest. For example, despite its diverse array of 13 biomes, supporting a wide range of biotic and abiotic habitats, only 3 species have been recorded from the southwestern state of Arizona: *Paragordius varius* (Leidy, 1851), *Gordionus violaceus* (Baird, 1853) and *Pseudochordodes gordioides* (Montgomery, 1898) as well as a yet to be described and named *Gordius* sp. from the Chiricahua mountains, part of the Sky Islands (Hanelt et al. 2015). We believe that this depauperate biodiversity is due to lack of study, and we have focused on investigating the northern tip of the Madrean Sky Island chain in southern Arizona.

#### **Methods**

#### Field collections

First field collections occurred on 27 July, 2011, at a stream in the Huachuca Mountains, Sunnyside, Cochise Country, Arizona, USA (31.445, -110.402, elevation: 1770 m). Subsequent collections were made on 28 July, 2011, from stream puddles near mile marker 12 on Madera Canyon Road, Madera Canyon, Santa Rita Mountains, Santa Cruz County, Arizona, USA (31.713, -110.87, elevation: 1640 m). All specimens were collected as free-living adults and transported alive, in stream water, to the laboratory. In the laboratory and before adult worms were processed for morphological and molecular analyses, worms were allowed to mate and females were allowed to deposit egg strings.

#### **Biological material and microscopy**

**Adults**. Physical attributes of specimen length and color were recorded in the laboratory. Measurements were obtained by placing specimens on a metric ruler, taking precautions to not stretch specimen. Specimens were cut into four pieces using razor blades. Pieces from the anterior, posterior, and mid-section were preserved in 70 % ethanol at room

temperature for future microscopy work. The remaining mid-section pieces were preserved in 100 % ethanol at -80 °C for future molecular analysis. Tissue samples preserved for microscopy were imaged using a Scanning Electron Microscope (SEM). Specimens were cleaned of debris using a previously-described method (Salas et al. 2011). Briefly, two drops of Clinique make up remover (Clinique, New York City, New York) placed into 1.5 ml tubes containing 70 % ethanol. The 1.5 ml tubes were placed into iSonic<sup>®</sup> Ultrasonic Cleaner Model D7810A (iSonic Inc., Chicago, Illinois), and cleaned for 4 minutes at maximum speed. Specimens were prepared by placing them in four increasing concentrations of ethanol (70 %, 85 %, 95 %, 100 %). Specimens were then dehydrated by placing them in increasing concentrations of hexamethyldisilazane (HMDS). Tissues were then mounted on stubs with carbon tape and coated with gold-palladium in an EmiTech K950 turbo-pumped vacuum coater with the gold-palladium sputter coater attachment (Quorum Technologies, West Sussex, England). Observations were made and digital images were taken using a JEOL 5800LV SEM at 15 kV (JEOL Ltd., Tokyo, Japan).

**Eggs and larvae using light microscopy.** For egg and larval measurements, pieces of egg string and hatched larvae were prepared as live wet mounts and observed using an Olympus BX-51 upright research microscope configured for bright field and DIC microscopy with plain fluorite objectives at 400× to 1000× total magnification. For egg measurements, the length and width was recorded for 30 eggs. For larvae, the length and width of the preseptum, postseptum, pseudointestine and stylets was measured for 30 larvae following the protocols of Szmygiel et al. (2014). Measurements of egg and larval characteristics were taken by capturing digital images of eggs and larvae using an Olympus 5 megapixel digital camera and ImageJ software to obtain measurements (Schneider et al. 2012). In addition, the morphology of the psuedointestine was recorded for larvae.

Larval preparation for SEM and external larval characteristics. Poly-L-Lysine coated cover-slips were placed in 1.5 ml plastic well plates. Frozen and live larvae were then thawed, suspended in water, then pipetted onto the Poly-L-Lysine coated coverslips and fixed in 10 % neutral buffered formalin. Poly-L-Lysine coated cover-slips with fixed larvae were dehydrated in a graded series of ethanol by first placing the Poly-L-Lysine coated cover-slip with fixed larvae in a 1.5 ml plastic well with 0.5 ml of 30 %, 50 % and 70 % ethanol for 30 min each. Next, 1 ml of 100 % ethanol was dripped into the well over a period of an hour, 1 ml of ethanol was then removed from the well and the process repeated 3 additional times. Finally, specimens were dried using HDMS, mounted on aluminum stubs, coated with gold palladium, and examined with a FEI Quanta 600 field emission gun ESEM (ThermoFisher Scientific, Hillsboro, OR) with Evex EDS and HKL EBSD as described previously (Szmygiel et al. 2014). The following morphological surface characteristics were recorded for at least 30 individual larvae: number of terminal spines on the postseptum, the number and relative size of cuticular hooks on the preseptum, the proboscis orientation (dorso-ventrally or laterally compressed) and the number and orientation of spines on the proboscis. Morphological characteristics for larvae examined with SEM followed terminology by Szmygiel et al. (2014).

**Infection of snails to obtain cysts.** Hatched larvae were collected with a Pasteur pipette and approximately 100 larvae were pipetted into 48 1.5 ml well plates filled with 1 mm of aged tap water. Four species of laboratory reared snails from three families maintained at Oklahoma State University following the protocol of Gustafson et al. (2015) were used for infections. Snail species included Physa (Physella) gyrina (Say, 1821), Stagnicola elodes (Say, 1821), the M line of Biomphalaria glabrata (Say, 1818), and Planorbella trivolvis (Say, 1817). For each snail species, 10 individuals were used for exposures, and a single laboratory reared snail was added to each well. Snails fed on the larva mixture for 48 hrs, were then removed and maintained based on species in 3.75 L jars filled with aerated aged tap water with a calcium gravel substrate. Snails were fed on a diet of frozen lettuce and Tetra Min® fish food and gordiid cysts were allowed to develop over a period of four weeks post exposure. Every seven days post exposure (DPE) a few individuals of each snail species were placed in labeled and capped 50 ml plastic centrifuged tubes, filled with approximately 35 ml of aged tap water, and frozen at -80 °C following the protocol of Bolek et al. (2013). Before dissection, centrifuge tubes were thawed and all snails were removed. Snails were processed for gordiid cysts following Harkins et al. (2016). Briefly, the snail body was removed with forceps from its shell under a dissection microscope and then pressed between two slides. Once snail tissue was flattened, a wet mount was prepared by removing the top slide and adding a drop of water and covering the flattened tissue with a coverslip. Slides were then examined with an Olympus BX-51 upright research microscope (Olympus, Tokyo, Japan) configured for bright field and differential interference contrast microscopy with plain fluorite objectives and a calibrated ocular micrometer at 100× to 400× total magnification. The status and degree of infection were determined by scanning the entire flattened snail carcass for cysts at 100× to 400× total magnification. Twenty cysts were digitally photographed at 1000× total magnification and the length and width of the cyst, cyst wall and encysted larvae were obtained using ImageJ software (Schneider et al. 2012) as previously described for larvae. Morphological characteristics for cysts followed terminology by Harkins et al. (2016).

**Infection of crickets to obtain adults.** Since *P. varius* and *P. obamai* can be domesticated by use of *Acheta domesticus* crickets as definitive hosts (Hanelt et al. 2012; Hanelt and Janovy 2004b), we experimentally-exposed *A. domesticus* to *P. amicus* sp. n. cysts from *Physa acuta* snails. Methods as outlined in Hanelt et al. 2012 were followed.

#### Molecular methods

A 1.0 cm mid-section piece, approximately 0.5–2.0 g, was cut into small pieces, dried at room temperature, and DNA was extracted using the E.Z.N.A. Mollusc DNA Kit (Omega Bio-Tek, Norcross, Georgia), following manufacturer instructions. DNA yield was determined using a NANO DROP 2000c spectrophotometer (Thermo Scientific, Walthem, MA). The *Paragordius* specific cytochrome c oxidase I (CO1) gene was amplified using modified universal CO1 primers (Folmer et al. 1994) Paragordius\_cox1F: GGT TAT AGA AAT ACA CAC TCC ATC TT and Paragordius\_cox1R2: TAA ACT TCA GGA TGA CCC AAA AAA CC. Subsequent PCR reactions used GoTaq Flexi DNA Polymerase (Promega Corp., Madison, Wisconsin) and were done following

manufacturer's instructions. Agarose gel electrophoresis was done using 1.0 % agarose gels, stained with 0.5 % GelRed Nucleic Acid Gel Stain (Biotium, Hayward, California), and visualized for bands on a UV transilluminator. Amplicons were purified by ethanol precipitation and sequenced using the BigDye version 3.1 kit (Applied Biosystems, Foster City, California) on an ABI 3130× sequence analyzer (Applied Biosystems). Both strands of the sequenced DNA fragments were assembled and edited in Sequencer version 5.0 (Gene Codes, Ann Arbor, Michigan).

# Molecular analyses

Partial CO1 sequences were aligned by eye; no sequences contained indels. As outgroups, two previously-published sequences from *Paragordius* spp. (Table 1) were included, as well as *P. varius* samples from across the United States. Evolutionary history was inferred by using the maximum likelihood method based on the Kimura 2-parameter (K2P) model (Kimura 1980) in MEGA7 (Kumar et al. 2016). All positions containing missing data were eliminated leaving a total of 437 positions in the final dataset. Internal support was assessed using 1,000 bootstrap replicates. CO1 genetic distances between each pair of samples were calculated using the K2P model in MEGA7. This dataset included 658 base pairs. Data were summarized for within and between genetic groups.

Species/sample	Accession <sup>†</sup>	Collection location <sup>‡</sup>	Lat.	Long.	Genbank accession			
Paragordius amicus sp. n.								
N289A	MSB:Para:26389	Arizona	31.713	-110.874	MG654049			
N289B	MSB:Para:26390	Arizona	31.713	-110.874	MG654050			
N291A	MSB:Para:26387	Arizona	31.445	-110.402	MG654047			
N291B	MSB:Para:26388	Arizona	31.445	-110.402	MG654048			
Paragordius varius								
N000	MSB:Para:26391	Nebraska	40.994	-96.566	MG654052			
N138 <sup>§</sup>	MSB:Para:26392	Montana			MG654053			
N210	MSB:Para:26393	Missouri	37.300	-89.550	MG654054			
N256	MSB:Para:26394	Mississppi	34.359	-88.462	MG654055			
N364A	MSB:Para:26395	New Mexico	34.766	-106.328	MG654056			
N364B	MSB:Para:26396	New Mexico	34.766	-106.328	MG654057			
N398		Oklahoma			KU721073			
Outgroups								
<i>Paragordius</i> sp. <sup>§</sup>		South Africa			AY428843			
Paragordius obamai	MSB:Para:26397	Kenya	-0.152	34.446	MG654059			
Paragordius tricuspidatus	MSB:Para:26398	France	43.755	3.110	MG654058			

Table 1. Collecting location for hairworm samples used in this study.

<sup>†</sup> Museum of Southwestern Biology, Parasitology Division.

<sup>‡</sup> Within the United States unless otherwise noted.

§ Exact locality is unknown.

#### Results

#### Taxonomy

#### Paragordius amicus sp. n.

http://zoobank.org/6C823753-BBBB-4749-B29F-31FFCF4ED784

**Type locality.** Huachuca Mountains, Sunnyside, Arizona, USA (31.445, -110.402, elevation: 1770 m).

**Holotype.** Female collected on 27 July, 2011, from type locality (N291A). Deposited into the Museum of Southwestern Biology (MSB) Parasite Division, University of New Mexico (UNM), New Mexico, USA with accession number MSB:Para:26387.

**Paratypes.** Allotype: male specimen collected on 27 July, 2011, from the type locality (N291B). Deposited into the MSB Parasite Division, accession number MSB:Para:26388. Paratypes: two females collected 28 July 2011, in the Santa Rita Mountains (N289A, and N289B). Deposited into the MSB Parasite Division, accession numbers MSB:Para:26389 and MSB:Para:26390.

**Host.** Natural definitive insect host is unknown; in the laboratory, *Acheta domesticus*, crickets served as definitive hosts, but in nature are likely to be members within the family Gryllidae (crickets) or Tettigoniidae (bush-crickets or katydids).

**Etymology.** The name is Latin for "friend", referring to the fact this is the first description of another genus member for *P. varius* in the Nearctic.

**Distribution.** Current known distribution is limited to the Madrean Sky Islands of southeastern Arizona in the Huachuca and Santa Rita Mountain Ranges.

**Material examined.** Adults (n=5), eggs, larvae, and cysts. Tissue from field collected adult (N=4) midsections was utilized for CO1 analysis while adult posterior, anterior, and midsections were utilized for SEM. DNA was also extracted from a worm removed from a deceased, lab-infected *Acheta domesticus* 40 days post exposure to collected larvae. Egg, larvae, and cyst stages were imaged using SEM and/or DIC microscopy.

**Description of male.** Adult (n=1) 205 mm long medium brown color. Bifurcating tail lobes on posterior roughly 400  $\mu$ m in length, extending laterally away from the sagittal plane (Fig. 1B). Male cloacal opening oval with a vertical slit-like opening (30×60  $\mu$ m, located anterior to point of tail lobe bifurcation (Fig. 1C). Post-cloacal spines present just above bifurcation but posterior to the cloaca extending onto the inside and ventral side of the tail lobes (Fig. 1B, C). Midbody cuticle lacks of obvious surface structures; some superficial structure is noted as dark-appearing areas (Fig. 1C). Cuticle on posterior end has wrinkled appearance made of grooves and circular pattern and is evenly dispersed across the cuticle surface with the exception of a 25  $\mu$ m wide line running on the ventral line of cuticle is bordered by small pointed mounds approximately the same size and shape as postcloacal spines but are mound rounded and mound-like (Fig. 1A, C).

**Description of female.** Adults (n=3) were 198 mm, 216 mm, and 234 mm in length and medium brown in color. Trifurcated posterior end (Figs 2C, 3A, 4A), with varying



**Figure 1.** *Paragordius amicus* sp. n. adult male from the Huachuca Mountains. **A** Midbody cuticle showing lack of obvious surface structure; some superficial structure is seen (arrows) **B** Bifurcating posterior end exhibiting the characteristic male bifurcating lobes with small, circular pointed lobes extending from the base of the bifurcation up the interior side of the lobes. These mounds are also found on either side as well as below the cloacal opening, eventually merging into the border of the ventral smooth cuticle line. **C** Cloacal opening, oblong with a straight slit opening extending the length of the opening surrounded by spines.

degrees of openness. Distinctive oblong mounds, approximately 10µm in length, are found arranged in horizontal or vertical lines up and down the entire interior side of the trifurcating tail lobes (Figs 2C, D, 3A, B). Short, thick bristles (hair-like structures) randomly spaced between oval mounds (Figs 2D, 3B). Midbody cuticle geographically variable. Worms collected in the Santa Rita Mountains contained transverse striations consisting of raised ridges separated by narrow furrows (Figs 2A, B, 4B, C, D). In some areas, the cuticle also contains rounded indentations (Figs 2A, 4B, C), while in others the indentations were more oblong and housed structures (Figs 2B, 4D). The female collected from the Huachuca Mountains, just as the male collected from the Huachuca Mountains, lacked any obvious surface structure on the midbody cuticle (Figs 1A, 3C).

**Description of egg strings, eggs, and larvae.** Females deposited continuous egg strings that were white in color and 1–3 times the length of the females. Eggs were elliptical to spherical in shape with a thin shell and were  $36.6 (29.6-41.2) \mu m$  in length and  $32.0 (25.9-43.8) \mu m$  in width. Over a period of 3–4 weeks, egg strings turned a light brown color at which time eggs contained fully developed larvae (Fig 5A).



**Figure 2.** *Paragordius amicus* sp. n. adult female from the Santa Rita Mountains **A–B** Midbody cuticle exhibiting transverse striations made of ridges separated by furrows **A** In some areas with hollow round to oblong indentations **B** In other areas with more oblong indentations containing structures **C** Trifurcating posterior. Note the cuticle features that can be seen on the interior surface of the lobes **D** Increased magnification of the lobes showing the oblong mounds (arrows). Note the long and thin hairs extending from some of the furrows between the mounds.

Larvae possessed a cylindrical body divided by a septum into two regions, the preseptum and a postseptum (Fig. 5B, C). The preseptum was 29.4 (24–38)  $\mu$ m in length and 15.4 (13–17)  $\mu$ m in width and contained an eversible proboscis supported by three internal stylets which were 13.8 (12–16)  $\mu$ m in length and 4.5 (3–6)  $\mu$ m in width (Fig. 5A, B). The postseptum was 34.8 (29–39)  $\mu$ m in length and 12.6 (10–15)  $\mu$ m in width and contained a clearly visible pseudointestine. The pseudointestine contained two anterior granules and a large posterior mass and was 15.5 (11–18)  $\mu$ m in length and 10.1 (7–13)  $\mu$ m in width (Fig. 5B). The average preseptum to post septum ratio was 1:1.2 (1.1–1.5).

Externally, larvae were superficially annulated and the postseptum contained two pairs of terminal spines located ventrally (Fig. 5C, E). The preseptum contained three sets of cuticular hooks. The outer ring of hooks contained seven hooks, two of which are very close together and ventrally positioned (Fig. 5D), and there were six hooks in the middle and inner rings observed in live larvae. The length of the cuticular hooks on the outer ring was noticeably longer than the middle and inner cuticular hooks. Clearly



**Figure 3**. *Paragordius amicus* sp. n. adult female from the Huachuca Mountains **A** Trifurcating posterior. **B** High magnification view of the oblong mounds and long, thin hair extensions found on the interior surface of the trifurcating lobes **C** Midbody cuticle lacking obvious surface structure.

visible spines on the proboscis could only be observed in a few individuals (Fig. 5D). The left and right side of the distal end of the dorsoventrally compressed and eversible proboscis each contained spines (at least four pairs arranged in tandem and one single spine above); whereas the distal end of the ventral side of the proboscis contained five spines (two pairs arranged in tandem and one single spine above; Fig 5D).

**Laboratory rearing of cysts and adults, and description of cyst.** Of the four snail species exposed to larvae of *P. amicus* sp. n. only *Physa acuta* and *Biomphalaria glabrata* became infected with cysts; however, not all individuals became infected. Seven of 10 (70 %) *P. acuta* were infected with a mean abundance of  $2.8 \pm 3.0$  (range 0–8) cysts; and four of 10 (40 %) of *B. glabrata* were infected with a mean abundance of  $1.0 \pm 2.2$  (0–7) cysts.

Fully developed cysts were recovered from laboratory-reared and exposed snails 14– 21 DPE. They contained a clear cyst wall of unknown composition 16.1 (12–24)  $\mu$ m in length and 11.5 (9–13)  $\mu$ m in width (Fig. 5F). During cyst formation the content of the larval pseudointestine was emptied and the larva folded its postseptum twice around the preseptum. However, unlike cysts of other gordiid genera, the posterior end of the postseptum never reached the posterior end of the preseptum (Fig 5F). The folded larva inside of the cyst was 28.1 (26–29)  $\mu$ m in length and 18.6 (18–20)  $\mu$ m in width.



**Figure 4.** *Paragordius amicus* sp. n. adult female from the Santa Rita Mountains **A** Trifurcating posterior **B–D** Midbody cuticle surface of with transverse striations containing round to slightly oblong indentations **B–C** Areas with only hollow indentations **D** Area of cuticle with more oblong indentations housing structures.

Of approximately 40 *A. domesticus* crickets exposed to about 10–100 cysts each, approximately 7 worms developed in 4 cricket hosts. To establish that the parasite infection was *P. amicus* sp. n. one worm was extracted for DNA, amplified, and sequenced as described above. The sequence was 100 % identical to both worms sequenced from the Santa Rita Mountains, and was placed into Genbank as MG654051.

Diagnosis and taxonomic comments. Paragordius amicus sp. n. has unique morphological features which warrant placing it as a new species and make it distinct from other New World Paragordius. First, the semi-oval raised cuticle structures and the short bristles (hair-like structures) on the inside of the female tail lobes have not been documented previously in Paragordius species. Second, despite the geographical variation in the cuticle structure of *P. amicus* sp. n., both variants have a cuticle pattern not seen in Nearctic and New World Paragordius species. Paragordius varius is the only species in the Americas also containing transverse striations separated by furrows. However, in *P. varius* the ridges within the striations are topped with round knobs (Schmidt-Rhaesa et al. 2003). Paragordius flavescens Linstow, 1906, found in South America, and *P. diversolobatus* Heinze, 1935 from Costa Rica contain areoles. Paragordius esavianus Carvalho, 1942, from South America lacks areoles but the cuticle is covered by dispersed round tubercles, longer bristles, and irregular small cuticular



**Figure 5.** Egg, larva and cyst characteristics of *Paragordius amicus* sp. n. **A** DIC photomicrograph of an egg with a fully developed larva. Note the stylets (arrow) **B** DIC photomicrograph of a live larva. Note the pseudointestine composed of two anterior granules (gr) and posterior mass (pm) **C** SEM photomicrograph of larva. Note the distinct preseptum (pre) with relatively long outer hooks, and postseptum (pos) with posterior spines **D** SEM photomicrograph of the anterior end larva. Note the partially everted proboscis (p) with distinct spines on the left and right lateral sides (white arrows), and the outer row of hooks containing two ventral outer hooks (voh) **E** SEM photomicrograph of larva. Note two posterior spines (ps) on the dorsal side of the postseptum (pos) **F** DIC photomicrograph of cyst (dorso-ventral view). Note the clear cyst wall (white arrow), distinct spines on the preseptum (black arrows), and the posterior end of the postseptum (white arrow).

elevations. *Paragordius minusculus* Carvalho, 1944, found in Brazil, lacks areoles but the midbody cuticle is fully covered by digit-like cuticular projections, like bristles, with blunt apexes. Finally, *Paragordius andreasii*, Zanca & de Villalobos, 2006, from Argentina, has a midbody cuticle with oval or rounded depressions arranged in pairs or forming perpendicular lines to the axis of the body. Finally, the female *P. obamai* Hanelt et al., 2012, does contain structures on the inside of the tail lobes. However, these structures are longitudinal, parallel ridges from which more narrow and longer bristles emerge (Hanelt et al. 2012).

Morphological characteristics of egg stings, eggs, larvae, and cysts of *Paragordius amicus* sp. n. were indistinguishable from these non-adult stages of two other species of *Paragordius* (*P. obamai* and *P. varius*) for which non-adult descriptions exist (Szmygiel et al. 2014). However, egg strings, larvae, and cysts of *Paragordius amicus* sp. n. were



**Figure 6**. Relationships inferred by maximum likelihood. Tree is unrooted, and drawn to scale indicating number of substitutions per site. Bootstrap values above 0.90 are shown.

morphologically distinct from egg strings, larvae of other gordiid genera such as *Acuto-gordius*, *Chordodes*, *Gordius* and *Neochordodes* (Chiu et al. 2017; Szmygiel et al. 2014).

**Molecular data.** Genetic distances of the CO1 barcoding region supports our contention that *P. amicus* sp. n. is a new species and that it is distinct from *P. varius*. The intraspecific distances among *P. varius* samples from around the USA is 0.72 %, while among *P. amicus* sp. n. samples is 1.09 %. The interspecific distance between *P. varius* and *P. amicus* sp. n. is 25.33 %. The inferred phylogenetic relationship (Fig. 6) supports the clustering of *P. varius* from around the USA forming a monophyletic group.

# Discussion

*Paragordius amicus* sp. n. represents the first hairworm described as an endemic to the Madrean Sky Islands, and so far only the second new species to be documented form the Chiricahua Mountains (see also Hanelt et al. 2015). There has been increased need for diversity studies like this in light of the emerging global crisis of climate change, especially for parasites which are chronically understudied organisms (Carlson et al. 2017; Dougherty et al. 2016). Climate change is considered the greatest threat to biodiversity, and its affects are often disproportionate depending upon an organism's life history, distribution, and location (Dunn et al. 2009; Malcolm et al. 2006). Due to their dependence on aquatic and terrestrial habitat and the spatial and temporal synchronization of paratenic and definitive hosts, hairworms may be a group heavily impacted by the effects of climate change.

The Madrean Sky Islands are considered a biodiversity hotspot, or "cradles of diversity", and thus climate change may have a proportionally large impact on this ecosystem (Malcolm et al. 2006; McCormack et al. 2009; O'Neal et al. 2005). The Sky Islands are actually expected to experience the influences associated with climate change sooner than other parts of the west (Garfin et al. 2013; Notaro et al. 2012). The most immediate of these impacts is wildfire. In the Western U.S.A, the average annual area affected by wildfires has increased by more than six fold over the past four decades (Littell et al. 2009; Westerling et al. 2006). Indeed, since collecting the specimens described in this study, several fires have swept both the Santa Rita Mountains (e.g. Sawmill fire, April 2017, 47,000 acres) and the Huachuca Mountains (e.g. Monument fire, June 2011, 29,000 acres).

Like oceanic islands, Sky Islands are habitat surrounded by barriers to biological dispersal. These barriers lead to isolation and ultimately high rates of endemism. Although *Paragordius amicus* sp. n. is not isolated to a single island, we have tantalizing evidence that the populations on the Santa Rita Mountains and Huachuca Mountains, separated by just 53.5 kilometers, may have been temporally isolated. These two populations appear to vary morphologically, in their cuticle pattern, but also are separated genetically by about 1.1%. Comparatively, *P. varius*, collected from several locations separated by hundreds and up to 1,600 kilometers apart vary genetically by only an average of 0.72%. In the future, we hope to collect additional specimens to more thoroughly document geographical variation in morphology and genetics.

#### Acknowledgements

This work was supported by the National Science Foundation, award numbers DEB-0949951 to Matthew G. Bolek and DEB-0950066 to Ben Hanelt and Andreas Schmidt-Rhaesa. We thank Terry Colberg and Lisa Whitworth of the OSU microscopy facility and Michael Spilde of the UNM Earth and Planetary Sciences SEM facility for their invaluable help with SEM work during this study. In addition, we thank Ryan Shannon, Chelsie Pierce, and Kristen Enyart for help in infecting and maintaining snails. We would also like to thank the UNM Center for Evolutionary and Theoretical Immunology (CETI) and the UNM Molecular Biology Facility, supported by a National Institute of Health grant 1P20RR18754 from the Institute Development Award Program of the National Center for Research Resources. Finally, we would like to thank two Citizen Scientists who collected and sent samples to us. Ryan Elwell sent worms from Missouri, and Maggie Oswald, at age 11, sent us samples from Mississippi.

#### References

Biron DG, Joly C, Menigoz A, Hanelt B, Thomas F (2005) Water seeking behaviour in insects harbouring hairworms: should the host collaborate with the parasite? Behavioral Ecology 16: 656–660. https://doi.org/10.1093/beheco/ari039

- Bolek MG, Rogers E, Szmygiel C, Shannon RP, Doerfert-Schrader WE, Schmidt-Rhaesa A, Hanelt B (2013) Survival of larval and cyst stages of Gordiids (Nematomorpha) after exposure to freezing. Journal of Parasitology 99: 397–402. https://doi.org/10.1645/12-62.1
- Carlson CJ, Burgio KR, Dougherty ER, Phillips AJ, Bueno VM, Clements CF, Castaldo G, Dallas TA, Cizauskas CA, Cumming GS, Dona J, Harris NC, Jovani R, Mironov S, Muellerklein OC, Proctor HC, Getz WM (2017) Parasite biodiversity faces extinction and redistribution in a changing climate. Science advances 3: e1602422. https://doi.org/10.1126/sciadv.1602422
- Chiu MC, Huang CG, Wu WJ, Shiao SF (2017) A new orthopteran-parasitizing horsehair worm, *Acutogordius taiwanensis* sp. n., with a redescription of *Chordodes formosanus* and novel host records from Taiwan (Nematomorpha, Gordiida). 683: 1–23. https://doi. org/10.3897/zookeys.683.12673
- Dougherty ER, Carlson CJ, Bueno VM, Burgio KR, Cizauskas CA, Clements CF, Seidel DP, Harris NC (2016) Paradigms for parasite conservation. Conservation biology 30: 724–733. https://doi.org/10.1111/cobi.12634
- Dunn RR, Harris NC, Colwell RK, Koh LP, Sodhi NS (2009) The sixth mass coextinction: are most endangered species parasites and mutualists? Proceedings Biological Sciences 276: 3037–3045. https://doi.org/10.1098/rspb.2009.0413
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Molecular Marine Biology and Biotechnology 3: 294–299.
- Garfin G, Jardine A, Merideth R, Black M, LeRoy S (2013) Assessment of climate change in the Southwest United States: A report prepared for the National Climate Assessment. Island Press, Washington, DC, 506 pp. https://doi.org/10.5822/978-1-61091-484-0
- Gustafson KD, Belden JB, Bolek MG (2015) The effects of the herbicide atrazine on freshwater snails. Ecotoxicology 24: 1183–1197. https://doi.org/10.1007/s10646-015-1469-x
- Hanelt B, Bolek MG, Schmidt-Rhaesa A (2012) Going solo: discovery of the first parthenogenetic gordiid (Nematomorpha: Gordiida). PLoS One 7: e34472. https://doi.org/10.1371/ journal.pone.0034472
- Hanelt B, Janovy J Jr. (2004a) Life cycle and paratenesis of American Gordiids (Nematomorpha: Gordiida). Journal of Parasitology 90: 240–244. https://doi.org/10.1645/GE-78R
- Hanelt B, Janovy J Jr. (2004b) Untying a Gordian knot: the domestication and laboratory maintenance of a Gordian worm, *Paragordius varius* (Nematomorpha: Gordiida). Journal of Natural History 39: 939–950. https://doi.org/10.1080/0022293021000058718
- Hanelt B, Schmidt-Rhaesa A, Bolek MG (2015) Cryptic species of hairworm parasites revealed by molecular data and crowdsourcing of specimen collections. Molecular Phylogenetics and Evolution 82: 211–218. https://doi.org/10.1016/j.ympev.2014.09.010
- Hanelt B, Thomas F, Schmidt-Rhaesa A (2005) Biology of the phylum Nematomorpha. Advances in Parasitology 59: 243–305. https://doi.org/10.1016/S0065-308X(05)59004-3
- Harkins C, Shannon R, Papes M, Schmidt-Rhaesa A, Hanelt B, Bolek MG (2016) Using Gordiid cysts to discover the hidden diversity, potential distribution, and new species of Gordiids (Phylum Nematomorpha). Zootaxa 4088: 515–530. https://doi.org/10.11646/ zootaxa.4088.4.3
- Kimura M (1980) A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. Journal of Molecular Evolution 16: 111–120. https://doi.org/10.1007/BF01731581
- Kumar S, Stecher G, Tamura K (2016) MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. Molecular Biology and Evolution 33: 1870–1874. https://doi. org/10.1093/molbev/msw054
- Littell JS, McKenzie D, Peterson DL, Westerling AL (2009) Climate and wildfire area burned in western U.S. ecoprovinces, 1916–2003. Ecological applications : a publication of the Ecological Society of America 19: 1003–1021. https://doi.org/10.1890/07-1183.1
- Malcolm JR, Liu C, Neilson RP, Hansen L, Hannah L (2006) Global warming and extinctions of endemic species from biodiversity hotspots. Conservation Biology 20: 538–548. https://doi.org/10.1111/j.1523-1739.2006.00364.x
- McCormack JE, Huateng H, Knowles LL (2009) Sky Islands. In: Gillespie RG, Clague DA (Eds) Encyclopedia of Islands. University of California Press, Berkeley, CA, 839–843.
- Notaro M, Mauss A, Williams JW (2012) Projected vegetation changes for the American Southwest: combined dynamic modeling and bioclimatic-envelope approach. Ecological applications: a publication of the Ecological Society of America 22: 1365–1388. https:// doi.org/10.1890/11-1269.1
- O'Neal KJ, Rogan J, Yool SR (2005) Monitoring post-fire vegetation regeneration in a Madrean ecosystem. USDA Forest Service Proceedings 36: 533–535.
- Poinar GO (2008) Global diversity of hairworms (Nematomorpha: Gordiaceae) in freshwater. Hydrobiologia 595: 79–83. https://doi.org/10.1007/s10750-007-9112-3
- Salas L, De Villalobos C, Zanca F (2011) Sexual size dimorphism, sex ratio and the relationship between seasonality and water quality in four species of Gordiida (Nematomorpha) from Catamarca, Argentina. Journal of Helminthology 85: 319–324. https://doi.org/10.1017/ S0022149X1000057X
- Schmidt-Rhaesa A, Hanelt B, Reeves WK (2003) Redescription and compilation of Nearctic freshwater Nematomorpha (Gordiida), with the description of two new species. Proceedings of the Academy of Natural Sciences of Philadelphia 153: 77–117. https://doi. org/10.1635/0097-3157(2003)153[0077:RACONF]2.0.CO;2
- Schneider CA, Rasband WS, Eliceiri KW (2012) NIH Image to ImageJ: 25 years of image analysis. Nature Methods 9: 671–675. https://doi.org/10.1038/nmeth.2089
- Szmygiel C, Schmidt-Rhaesa A, Hanelt B, Bolek MG (2014) Comparative descriptions of non-adult stages of four genera of Gordiids (Phylum: Nematomorpha). Zootaxa 3768: 101–118. https://doi.org/10.11646/zootaxa.3768.2.1
- Westerling AL, Hidalgo HG, Cayan DR, Swetnam TW (2006) Warming and earlier spring increase western U.S. forest wildfire activity. Science 313: 940–943. https://doi.org/10.1126/ science.1128834

CORRIGENDA



## Corrigenda: De Mattia W, Neiber MT, Groh K (2018) Revision of the genus-group Hystricella R. T. Lowe, 1855 from Porto Santo (Madeira Archipelago), with descriptions of new recent and fossil taxa (Gastropoda, Helicoidea, Geomitridae). ZooKeys 732: 1–125. https://doi.org/10.3897/zookeys.732.21677

Willy De Mattia<sup>1,2</sup>, Marco T. Neiber<sup>3</sup>, Klaus Groh<sup>4</sup>

I Natural History Museum Vienna, Burgring 7, 1010 Vienna, Austria 2 International Centre for Genetic Engineering and Biotechnology, Padriciano 99, 34149, Trieste, Italy 3 Centre of Natural History, Zoological Museum, University of Hamburg, Martin-Luther-King-Platz 3, D-20146 Hamburg, Germany 4 Hinterbergstraße 15, D-67098 Bad Dürkheim, Germany

Corresponding author: Willy De Mattia (demattia@icgeb.org)

Academic editor: M. Haase   Received 26 January 2018   Accepted 29 January 2018   1	Published 1 February 2018
- http://zoobank.org/59C124B1-8E0C-4372-A9A3-4F9823A07688	

**Citation:** De Mattia W, Neiber MT, Groh K (2018) Corrigenda: De Mattia W, Neiber MT, Groh K (2018) Revision of the genus-group *Hystricella* R. T. Lowe, 1855 from Porto Santo (Madeira Archipelago), with descriptions of new recent and fossil taxa (Gastropoda, Helicoidea, Geomitridae). ZooKeys 732: 1–125. https://doi.org/10.3897/zookeys.732.21677. ZooKeys 733: 147–148. https://doi.org/10.3897/zookeys.733.23906

We proposed the name *Wollastonia* De Mattia, Neiber & Groh, 2018 for a small group of land snails that are endemic to the island of Porto Santo and some of its sourrounding islet in the Madeiran Archipelago (De Mattia et al. 2018). Unfortunately, and only explicable as the consequence of a whole chain of small mistakes or omissions, it slipped our attention that the name is a junior homonym of *Wollastonia* Heer, 1852 (Coleoptera), *Wollastonia* Horn, 1873 (Coleoptera) and *Wollastonia* Machado, 1984 (Coleoptera) (see Heer 1852: 13, Horn 1873: 433–434, Machado 1984: 131). Consequently, the genus needs a new name and we herewith propose *Wollastonaria* nom. n. as a new replacement name for *Wollastonia* De Mattia, Neiber & Groh, 2018. The type species of *Wollastonaria* nom. n. is the nominal species *Helix* [*Helicella*] *turricula* R. T. Lowe, 1831. A detailed description of the genus is given in De Mattia et al. (2018) under the name *Wollastonia*. Included are the following species and subspecies:

Copyright Willy De Mattia et al. This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

*Wollastonaria turricula* (R. T. Lowe, 1831), comb. n., *Wollastonaria vermetiformis* (R. T. Lowe, 1855), comb. n., *Wollastonaria ripkeni* (De Mattia & Groh, 2018), comb. n., *Wollastonaria falknerorum* (Groh, Neiber & De Mattia, 2018), comb. n., *Wollastonaria leacockiana* (Wollaston, 1878), comb. n., *Wollastonaria beckmanni* (De Mattia & Groh, 2018), comb. n., *Wollastonaria jessicae jessicae* (De Mattia, Neiber & Groh, 2018), comb. n., *Wollastonaria jessicae jessicae* (De Mattia, Neiber & Groh, 2018), comb. n., *Wollastonaria jessicae monticola* (De Mattia, Neiber & Groh, 2018), comb. n., *Wollastonaria klausgrohi* (De Mattia & Neiber, 2018), comb. n., *Wollastonaria klausgrohi* (De Mattia & Neiber, 2018), comb. n., *Wollastonaria subcarinulata* (Wollaston, 1878), comb. n., and *Wollastonaria inexpectata* (De Mattia & Groh, 2018), comb. n.

Fig. 15 should read *Caseolus (Leptostictea) leptosticus*, Ponta do Garajau, Madeira. Fig. 16 should read *Caseolus (Helicomela) punctulatus punctulatus*, Fonte da Areia.

## Acknowledgements

We would like to thank Barna Páll-Gergely (Hungary) and Rob Oudejans (The Netherlands) who kindly pointed out to us that the proposed name is preoccupied.

## References

- De Mattia W, Neiber MT, Groh K (2018) Revision of the genus-group *Hystricella* R. T. Lowe, 1855 from Porto Santo (Madeira Archipelago), with descriptions of new recent and fossil taxa (Gastropoda: Helicoidea: Geomitridae). ZooKeys 732: 1–125. https://doi.org/10.3897/zookeys.732.21677
- Heer O (1852) Ueber die Lias–Insel im Aargau. In: Heer O, Escher von der Linth A (Eds) Zwei geologische Vorträge gehalten im März 1852. E. Kiesling, Zürich, 1–15. [1 pl.]
- Horn GH (1873) Contributions to a knowledge of the Curculionidae of the United States. Proceedings of the American Philosophical Society 13: 407–469.
- Machado A (1984) Pterostiquidos anoftalmos nuevos de las Islas Canarias y description de Wollastonia n. gen. (Coleoptera Caraboidea). La Nouvelle Revue d'Entomologie (Nouvelle Série) 1: 129–137.